

**PCT**WORLD INTELLECTUAL PROPERTY ORGANIZATION  
International Bureau

## INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

<b>(51) International Patent Classification <sup>6</sup> :</b> <b>C12N 9/42, C11D 3/386, D06M 16/00</b>	<b>A1</b>	<b>(11) International Publication Number:</b> <b>WO 99/57259</b> <b>(43) International Publication Date:</b> 11 November 1999 (11.11.99)
<b>(21) International Application Number:</b> PCT/US99/09409 <b>(22) International Filing Date:</b> 30 April 1999 (30.04.99) <b>(30) Priority Data:</b> PCT/US98/08904 1 May 1998 (01.05.98) US <b>(71) Applicant (for all designated States except US):</b> THE PROCTER & GAMBLE COMPANY [US/US]; One Procter & Gamble Plaza, Cincinnati, OH 45202 (US). <b>(72) Inventors; and</b> <b>(75) Inventors/Applicants (for US only):</b> SMETS, Johan [BE/BE]; Bollenberg 79, B-3210 Lubbeek (BE). BUSCH, Alfred [DE/BE]; Handelsstraat 210, B-1840 Londerzeel (BE). BAECK, Andre, Cesar [BE/BE]; Putsesteenweg 273, B-2820 Bonheiden (BE). BETTIOL, Jean-Luc, Philippe [FR/BE]; 93, avenue Slegers, B-1200 Brussels (BE). BOYER, Stanton, Lane [US/US]; 12 Fall River Court, Fairfield, OH 45014 (US). <b>(74) Agents:</b> REED, T., David et al.; The Procter & Gamble Company, 5299 Spring Grove Avenue, Cincinnati, OH 45217-1087 (US).		<b>(81) Designated States:</b> AE, AL, AM, AT, AT (Utility model), AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, CZ (Utility model), DE, DE (Utility model), DK, DK (Utility model), EE, EE (Utility model), ES, FI, FI (Utility model), GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SK (Utility model), SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).  <b>Published</b> With international search report.
<b>(54) Title:</b> LAUNDRY DETERGENT AND/OR FABRIC CARE COMPOSITIONS COMPRISING A MODIFIED CELLULASE		
<b>(57) Abstract</b>  The present invention relates to a modified enzyme which comprises a catalytically active amino acid sequence of a cellulolytic enzyme linked to an amino acid sequence comprising a Cellulose Binding Domain (CBD) having a relative binding constant (K <sub>r-a</sub> ) for binding to amorphous cellulose higher than 2.4/gcellulose, preferably higher than 3.5/gcellulose, more preferably higher than 4/gcellulose, for selective binding and hydrolysis of amorphous cellulose of cotton containing fabrics in a laundry and/or fabric care application. The present invention further relates to laundry detergent and/or fabric care compositions comprising this modified enzyme.		

10-12  
16117

**FOR THE PURPOSES OF INFORMATION ONLY**

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	ES	Spain	LS	Lesotho	SI	Slovenia
AM	Armenia	FI	Finland	LT	Lithuania	SK	Slovakia
AT	Austria	FR	France	LU	Luxembourg	SN	Senegal
AU	Australia	GA	Gabon	LV	Latvia	SZ	Swaziland
AZ	Azerbaijan	GB	United Kingdom	MC	Monaco	TD	Chad
BA	Bosnia and Herzegovina	GE	Georgia	MD	Republic of Moldova	TG	Togo
BB	Barbados	GH	Ghana	MG	Madagascar	TJ	Tajikistan
BE	Belgium	GN	Guinea	MK	The former Yugoslav Republic of Macedonia	TM	Turkmenistan
BF	Burkina Faso	GR	Greece	ML	Mali	TR	Turkey
BG	Bulgaria	HU	Hungary	MN	Mongolia	TT	Trinidad and Tobago
BJ	Benin	IE	Ireland	MR	Mauritania	UA	Ukraine
BR	Brazil	IL	Israel	MW	Malawi	UG	Uganda
BY	Belarus	IS	Iceland	MX	Mexico	US	United States of America
CA	Canada	IT	Italy	NE	Niger	UZ	Uzbekistan
CF	Central African Republic	JP	Japan	NL	Netherlands	VN	Viet Nam
CG	Congo	KE	Kenya	NO	Norway	YU	Yugoslavia
CH	Switzerland	KG	Kyrgyzstan	NZ	New Zealand	ZW	Zimbabwe
CI	Côte d'Ivoire	KP	Democratic People's Republic of Korea	PL	Poland		
CM	Cameroon	KR	Republic of Korea	PT	Portugal		
CN	China	KZ	Kazakhstan	RO	Romania		
CU	Cuba	LC	Saint Lucia	RU	Russian Federation		
CZ	Czech Republic	LJ	Liechtenstein	SD	Sudan		
DE	Germany	LK	Sri Lanka	SE	Sweden		
DK	Denmark	LR	Liberia	SG	Singapore		
EE	Estonia						

## **LAUNDRY DETERGENT AND/OR FABRIC CARE COMPOSITIONS COMPRISING A MODIFIED CELLULASE**

### **Field of the Invention**

The present invention relates to laundry detergent and/or fabric care compositions comprising a modified enzyme which comprises a catalytically active amino acid sequence of a cellulolytic enzyme linked to an amino acid sequence comprising a Cellulose Binding Domain (CBD) having a relative binding constant ( $K_r$ -a) for binding to amorphous cellulose higher than 2.4l/gcellulose, preferably higher than 3.5l/gcellulose, more preferably higher than 4 l/gcellulose.

### **Background of the invention**

Detergent compositions include nowadays a complex combination of active ingredients and in particular, detergent enzymes such as cellulases.

The activity of cellulase is one in which cellulosic fibres or substrates are attacked by the cellulase and is depending on the particular function of the cellulase, which can be endo- or exo- cellulase and on the respective hemicellulases. The cellulose structures are depolymerized or cleaved into smaller and thereby more soluble or dispersible fractions. This activity in particular on fabrics provides cleaning, rejuvenation, softening and generally improved handfeel characteristics to the fabric structure. This has been previously speculated to be the result of the cleavage of fibrils from the surface of fibres such that the main strand of the fibre becomes smoother, less available for incrustation, less likely to entangle with other fibers and optically less diffuse in its light reflecting and emission.

Improvements in the activity of particular cellulase compositions have been made over time in order to allow lower usage or shorter exposure times for similar benefits. In the detergent field, cellulases performing in a typical detergent wash environment are available with an activity at which the desired

cellulase performance is reached prior to the end of a wash-cycle. However, since the cellulase continues to react, even after having provided the desired performance, cleavage of the cellulose will continue. Therefore, there is a potential risk of tensile strength loss.

It should be noted however, that tensile strength loss of fabric is also an unavoidable result of mechanical action due to use/wearing and may further result from damage by a bleaching component in the laundry process, especially if the fabric is contaminated with metal compounds.

The object of the present invention is to provide laundry detergent and/or fabric care compositions which achieve excellent fabric care, including anti-wrinkle, anti-bobbling and anti-shrinkage properties to fabrics, as well as provide static control, fabric softness, colour appearance and fabric anti-wear properties and benefits, and which achieve excellent fabric cleaning while preventing tensile strength loss.

The above objective has been met by formulating laundry detergent and/or fabric care compositions comprising a modified enzyme which comprises a catalytically active amino acid sequence of a cellulolytic enzyme linked to an amino acid sequence comprising a Cellulose Binding Domain (CBD) having a relative binding constant ( $K_r$ -a) for binding to amorphous cellulose higher than 2.4l/gcellulose, preferably higher than 3.5l/gcellulose, more preferably higher than 4l/gcellulose.

WO91/10732 discloses an enzyme which exhibits cellulase activity, producible by a strain of *Bacillus* spp., NICMB 40250, especially an 75, 56 or 45 kD Molecular weight endoglucanase or characterised by a high specific activity for e.g. detergency application, providing softening, soil removal and colour clarification. Novel derivatives of cellulase enzymes combining a core region derived from said endoglucanase with a CBD derived from another cellulase enzyme or a combining a core region derived from another cellulase enzyme with a CBD derived from said endoglucanase, are therein contemplated to construct cellulase enzymes with improved binding properties.

WO94/07998 describes cellulase variants of a cellulase classified in family 45, comprising a CBD, a Catalytically Active Domain (CAD) and a region linking the

CBD to the CAD, wherein one or more amino acid residues have been added, deleted or substituted and/or another CBD is added at the opposite end of the CAD. These enzymes have improved properties such as alkaline activity, harshness reduction, compatibility with detergent ingredients, particulate soil removal, colour clarification, defuzzing, depilling, harshness reduction and sensitivity to anionic surfactants and peroxidase bleaching systems and are useful in detergent compositions.

WO95/16782 relates to the cloning and high level expression of novel truncated cellulase proteins or derivatives thereof in *Trichoderma longibrachiatum*. Combinations of different core regions with several CBDs are described in the fibre/fabric context.

A cellulolytic enzyme preparation comprising a cellulase with reduced mobility, e.g. by increasing the molecular weight or apparent size of the cellulase protein molecule or by insolubilising or immobilising the cellulase, has been disclosed in WO97/01629. The mobility of the cellulase component may be reduced by adsorption to an insoluble or soluble carrier e.g. via the existing or newly introduced CBD.

However, none of these documents disclose a modified enzyme which comprises a catalytically active amino acid sequence of a cellulolytic enzyme linked to an amino acid sequence comprising a Cellulose Binding Domain having a  $K_r$ -a of a definite value, thereby defining a cellulase especially adapted to selectively bind and hydrolyse amorphous cellulose of cotton containing fabrics for a laundry and/or fabric care application.

### **Summary of the invention**

The present invention relates a modified cellulolytic enzyme demonstrating selective binding and hydrolysis of amorphous cellulose in cotton containing fabrics. The present invention further relates to a laundry detergent and/or fabric care composition comprising said modified enzyme. Said compositions achieve excellent fabric care and fabric cleaning while preventing tensile strength loss. This modified enzyme comprises a catalytically active amino acid sequence of a

cellulolytic enzyme linked to an amino acid sequence comprising a Cellulose Binding Domain (CBD) having a relative binding constant (Kr-a) for binding to amorphous cellulose higher than 2.4l/gcellulose, preferably higher than 3.5l/gcellulose, more preferably higher than 4l/gcellulose.

In a further embodiment of the present invention, the present invention relates to a modified enzyme which comprises a catalytically active amino acid sequence of a cellulolytic enzyme linked to an amino acid sequence comprising a Cellulose Binding Domain further characterised by having a relative binding constant for bacterial microcrystalline cellulose (Kr-c) lower than 1l/gcellulose, preferably lower than 0.5l/gcellulose.

More preferably, the modified enzyme of the present invention, comprises a catalytically active amino acid sequence of a cellulolytic enzyme linked to an amino acid sequence comprising a Cellulose Binding Domain further characterised by having a relative binding constant (Kr-a) for binding to amorphous cellulose to a relative binding constant for bacterial microcrystalline cellulose (Kr-c) ratio above 3.

The present invention further relates to laundry detergent and/or fabric care compositions comprising said modified enzyme and which preferably further comprise a detergent ingredient selected from cationic surfactants, dye transfer inhibiting polymer, builders, - in particular zeolite A and sodium tripolyphosphate - and/or clays .

### **Detailed description of the invention**

The present invention relates to a modified enzyme which comprises a catalytically active amino acid sequence of a cellulolytic enzyme linked to an amino acid sequence comprising a cellulose binding domain having a relative binding constant for binding to amorphous cellulose higher than 2.4l/gcellulose, preferably higher than 3.5l/gcellulose, more preferably higher than 4 l/gcellulose.

#### **Cellulolytic enzyme**

A modified enzyme for use in accordance with the present invention comprises a catalytically active (enzymatically active) amino acid sequence (in general a polypeptide amino acid sequence) of a cellulolytic enzyme linked to an amino acid sequence comprising a Cellulose Binding Domain having a high affinity for binding to amorphous cellulose.

The catalytically active amino acid sequence in question may comprise or consist of the whole of - or substantially the whole of - the full amino acid sequence of the mature enzyme in question, or it may consist of a portion of the full sequence which retains substantially the same catalytic (enzymatic) properties as the full sequence.

Modified enzymes (enzyme hybrids) of the type in question, as well as detailed descriptions of the preparation and purification thereof, are known in the art [see, e.g., WO 90/00609, WO 94/24158 and WO 95/16782, as well as Greenwood et al., Biotechnology and Bioengineering 44 (1994) pp. 1295 - 1305]. The production of enzymes hybrid is described in WO 91/10732 wherein novel derivatives of cellulase enzymes combining a core region derived from a *Bacillus* NICB 40250 endoglucanase with a CBD derived from another cellulase enzyme or a combining a core region derived from another cellulase enzyme with a CBD derived from a *Bacillus* NICB 40250 endoglucanase, are constructed. WO 95/16782 describes the combinations of different core regions with several CBD and the cloning and high level expression of these novel truncated cellulase proteins or derivatives thereof, in *Trichoderma longibrachiatum*.

They may, e.g., be prepared by transforming into a host cell a DNA construct comprising at least a fragment of DNA encoding the cellulose-binding domain ligated, with or without a linker, to a DNA sequence encoding the enzyme of interest, and growing the transformed host cell to express the fused gene. One relevant, but non-limiting, type of recombinant product (enzyme hybrid) obtainable in this matter - often referred to in the art as a "fusion protein" - may be described by one of the following general formulae:

A-CBD-MR-X-B

### A-X-MR-CBD-B

In the latter formulae, CBD is an amino acid sequence comprising at least the cellulose-binding domain (CBD) *per se*.

MR (the middle region; a linking region) may be a bond, or a linking group comprising from 1 to about 100 amino acid residues, in particular of from 2 to 40 amino acid residues, e.g. from 2 to 15 amino acid residues. MR may, in principle, alternatively be a non-amino-acid linker (See below).

X is an amino acid sequence comprising the above-mentioned, catalytically (enzymatically) active sequence of amino acid residues of a polypeptide encoded by a DNA sequence encoding the cellulolytic enzyme of interest.

The moieties A and B are independently optional. When present, a moiety A or B constitutes a terminal extension of a CBD or X moiety, and normally comprises one or more amino acid residues.

It will thus, *inter alia*, be apparent from the above that a CBD in an enzyme hybrid of the type in question may be positioned C-terminally, N-terminally or internally in the enzyme hybrid. Correspondingly, an X moiety in an enzyme hybrid of the type in question may be positioned N-terminally, C-terminally, or internally in the enzyme hybrid.

Enzyme hybrids of interest in the context of the invention include enzyme hybrids which comprise more than one CBD, e.g. such that two or more CBDs are linked directly to each other, or are separated from one another by means of spacer or linker sequences (consisting typically of a sequence of amino acid residues of appropriate length). Two CBDs in an enzyme hybrid of the type in question may, for example, also be separated from one another by means of an -MR-X- moiety as defined above. One or more cellulose binding domain can be linked to the N-terminal and/or C-terminal parts of the cellulase core region. Any part of a CBD can be selected, modified, truncated etc.

Preferably, attention will be given in the construction of enzyme hybrids of the type in question to the stability towards proteolytic degradation. Two- and multi-domain proteins are particularly susceptible towards proteolytic cleavage of linker regions connecting the domains. Proteases causing such cleavage may, for example, be subtilisins, which are known to often exhibit broad substrate



specificities [see, e.g. : Grøn et al., Biochemistry 31 (1992), pp. 6011-6018; Teplyakov et al., Protein Engineering 5 (1992), pp. 413-420]. Glycosylation of linker residues in eukaryotes is one Nature's ways of preventing proteolytic degradation. Another is to employ amino acids which are less favoured by the surrounding proteases. The length of the linker also plays a role in relation to accessibility by proteases. Which "solution" is optimal depends on the environment in which the enzyme hybrid is to function. When constructing new enzyme hybrid molecules, preferably attention will be paid to the linker stability.

### Plasmids

Preparation of plasmids capable of expressing fusion proteins having the amino acid sequences derived from fragments of more than one polypeptide is well known in the art (see, for example, WO 90/00609 and WO 95/16782). The expression cassette may be included within a replication system for episomal maintenance in an appropriate cellular host or may be provided without a replication system, where it may become integrated into the host genome. The DNA may be introduced into the host in accordance with known techniques such as transformation, microinjection or the like.

Once the fused gene has been introduced into the appropriate host, the host may be grown to express the fused gene. Normally it is desirable additionally to add a signal sequence which provides for secretion of the fused gene. Typical examples of useful genes are :

- 1) Signal sequence – (pro-peptide) – carbohydrate-binding domain – linker – enzyme sequence of interest, or
  - 2) Signal sequence – (pro-peptide) – enzyme sequence of interest – linker – carbohydrate-binding domain,
- in which the pro-peptide sequence normally contains 5-100, e.g. 5-25, amino acid residues. The recombinant product may be glycosylated or non-glycosylated.

### Cellulolytic enzymes

Any enzyme which exhibits a cellulase activity is suitable for the purpose of the present invention and includes cellulases without naturally occurring CBD, cellulase to which a CBD of Kr-a for binding to amorphous cellulose higher than 2.4l/gcellulose, preferably higher than 3.5l/gcellulose, more preferably higher than 4 l/gcellulose, is added and/or cellulases having a CBD presenting a lower Kr value, to which a CBD of Kr-a higher than 2.4l/gcellulose, preferably higher than 3.5l/gcellulose, more preferably higher than 4 l/gcellulose, is then substituted or added.

The catalytically active amino acid sequence of a cellulolytic enzyme can be derived from any cellulase. The cellulases usable in the present invention include both bacterial or fungal cellulases. Preferably, they will have a pH optimum of between 5 and 12 and a specific activity above 50 CEVU/mg (Cellulose Viscosity Unit). Suitable cellulases are disclosed in U.S. Patent 4,435,307, Barbesgaard et al, J61078384 and WO96/02653 which discloses fungal cellulase produced respectively from *Humicola insolens*, *Trichoderma*, *Thielavia* and *Sporotrichum*. EP 739 982 describes cellulases isolated from novel *Bacillus* species. Suitable cellulases are also disclosed in GB-A-2.075.028; GB-A-2.095.275; DE-OS-2.247.832 and WO95/26398.

Examples of such cellulases are cellulases produced by a strain of *Humicola insolens* (*Humicola grisea* var. *thermoidea*), particularly the *Humicola* strain DSM 1800. Other suitable cellulases are cellulases originated from *Humicola insolens* having a molecular weight of about 50KDa, an isoelectric point of 5.5 and containing 415 amino acids; and a 43kD endoglucanase derived from *Humicola insolens*, DSM 1800, exhibiting cellulase activity; a preferred endoglucanase component has the amino acid sequence disclosed in PCT Patent Application No. WO 91/17243. Also suitable cellulases are the EGIII cellulases from *Trichoderma longibrachiatum* described in WO94/21801, Genencor, published September 29, 1994. Especially suitable cellulases are the cellulases having colour care benefits. Examples of such cellulases are cellulases described in European patent application No. 91202879.2, filed November 6, 1991 (Novo). Carezyme and Celluzyme (Novo Nordisk A/S) are especially useful. See also WO91/17244 and WO91/21801. Other suitable cellulases for fabric care and/or cleaning properties are described in WO96/34092, WO96/17994 and WO95/24471.

Preferred catalytically active amino acid sequences of a cellulolytic enzyme for the laundry detergent and/or fabric care compositions of the present invention are producible by a fungal strain, preferably produced by a strain of *Humicola insolens* and more preferably is a ~43kD endoglucanase derived from *Humicola insolens*, DSM 1800, exhibiting cellulase activity.

Said modified cellulolytic enzymes are normally incorporated in the laundry detergent and/or fabric care composition at levels from 0.0001% to 2%, preferably 0.0001% to 0.5%, more preferably 0.0005% to 0.1% of pure modified enzyme by weight of the detergent composition.

Preferred catalytically active amino acid sequence of a cellulolytic enzyme for specific applications are derived from alkaline cellulases, i.e. enzymes having an enzymatic activity of at least 10%, preferably at least 25%, more preferably at least 40% of their maximum activity at a pH ranging from 7 to 12, preferably 10.5. More preferred core cellulases are enzymes having their maximum activity at a pH ranging from 7 to 12, preferably 10.5.

The above-mentioned enzymes may be of any suitable origin, such as vegetable, animal, bacterial, fungal and yeast origin. Origin can further be mesophilic or extremophilic (psychrophilic, psychrotrophic, thermophilic, barophilic, alkalophilic, acidophilic, halophilic, etc.). Purified or non-purified forms of these enzymes may be used. Nowadays, it is common practice to modify wild-type enzymes via protein / genetic engineering techniques in order to optimise their performance efficiency in the laundry detergent and/or fabric care compositions of the invention. For example, the variants may be designed such that the compatibility of the enzyme to commonly encountered ingredients of such compositions is increased. Alternatively, the variant may be designed such that the optimal pH, bleach or chelant stability, catalytic activity and the like, of the enzyme variant is tailored to suit the particular cleaning application.

In particular, attention should be focused on amino acids sensitive to oxidation in the case of bleach stability and on surface charges for the surfactant compatibility. The isoelectric point of such enzymes may be modified by the substitution of some charged amino acids, e.g. an increase in isoelectric point

may help to improve compatibility with anionic surfactants. The stability of the enzymes may be further enhanced by the creation of e.g. additional salt bridges and enforcing metal binding sites to increase chelant stability.

## **Cellulose Binding Domain (CBD)**

### Cellulose Binding Domains

In the present context, the terms "amino acid sequence comprising a CBD, Cellulose Binding Domain or CBD" are intended to indicate an amino acid sequence capable of effecting binding of the cellulase to a cellulosic substrate (e.g., as described in P. Kraulis et al., Determination of the three-dimensional structure of the C terminal domain of cellobiohydrolase I from *Trichoderma reesei*. A study using nuclear magnetic resonance and hybrid distance geometry-dynamically simulated annealing. Biochemistry 28:7241-7257, 1989). The classification and properties of cellulose binding domains are presented in P. Tomme et al., in the symposium "Enzymatic degradation of insoluble polysaccharides" (ACS Symposium Series 618, edited by J.N. Saddler and M.H. Penner, ACS, 1995).

Cellulose-binding (and other carbohydrate-binding) domains are polypeptide amino acid sequences which occur as integral parts of large polypeptides or proteins consisting of two or more polypeptide amino acid sequence regions, especially in hydrolytic enzymes (hydrolases) which typically comprise a catalytic domain containing the active site for substrate hydrolysis and a carbohydrate-binding domain for binding to the carbohydrate substrate in question. Such enzymes can comprise more than one catalytic domain and one, two or three carbohydrate-binding domains, and they may further comprise one or more polypeptide amino acid sequence regions linking the carbohydrate-binding domain(s) with the catalytic domain(s), a region of the latter type usually being denoted a "linker".

Examples of hydrolytic enzymes comprising a cellulose-binding domain are cellulase, xylanases, mannanases, arabinofuranosidases, acetylesterases and chitinases. "Cellulose-binding domains" have also been found in algae, e.g. in

the red alga *porphyra purpurea* in the form of a non-hydrolytic polysaccharide-binding protein [see P. Tomme et al., Cellulose-binding domains - Classification and Properties in Enzymatic Degradation of Insoluble Carbohydrates, John N. Saddler and Michael H. Penner (Eds.), ACS Symposium Series, No. 618 (1996)]. However, most of the known CBDs (which are classified and referred to by P. Tomme et al. (*op. cit.*) as "cellulose-binding domains" derive from cellulases and xylanases.

In the present context, the term "cellulose-binding domain" is intended to be understood in the same manner as in the latter reference (P. Tomme et al., *op. cit.*) The P. Tomme et al. reference classifies more than 120 "cellulose-binding domains" into 10 families (I-X) which may have different functions or roles in connection with the mechanism of substrate binding. However, it is to be anticipated that new family representatives and additional families will appear in the future.

In proteins/polypeptides in which CBDs occur (e.g. enzymes, typically hydrolytic enzymes such as cellulases), a CBD may be located at the N or C terminus or at an internal position.

The part of a polypeptide or protein (e.g. hydrolytic enzyme) which constitutes a CBD *per se* typically consists of more than about 30 and less than about 250 amino acid residues. For example, those CBDs listed and classified in Family I in accordance with P. Tomme et al. (*op. cit.*) consist of 33-37 amino acid residues, those listed and classified in Family IIa consist of 95-108 amino acid residues, those listed and classified in Family VI consist of 85-92 amino acid residues, whilst one CBD (derived from a cellulase from *Clostridium thermocellum*) listed and classified in Family VII consists of 240 amino acid residues. Accordingly, the molecular weight of an amino acid sequence constituting a CBD *per se* will typically be in the range of from about 4kD to about 40kD, and usually below about 35kD.

Cellulose binding domains can be produced by recombinant techniques as described in H. Stålbrand et al., Applied and Environmental Microbiology, Mar. 1995, pp. 1090-1097; E. Brun et al., (1995) Eur. J. Biochem. 231, pp. 142-148; J.B. Coutinho et al., (1992) Molecular Microbiology 6(9), pp. 1243-1252

In order to isolate a cellulose binding domain of, e.g. a cellulase, several genetic engineering approaches may be used. One method uses restriction enzyme to remove a portion of the gene and then to fuse the remaining gene-vector fragment in frame to obtain a mutated gene that encodes a protein truncated for a particular gene fragment. Another method involves the use of exonucleases such as Ba131 to systematically delete nucleotides either externally from the 5' and the 3' ends of the DNA or internally from a restricted gap within the gene. These gene-deletion methods result in a mutated gene encoding a shortened gene molecule whose expression product may then be evaluated for substrate-binding (e.g. cellulose-binding) ability. Appropriate substrates for evaluating the binding ability include cellulosic materials such as Avicel <sup>TM</sup> and cotton fibres. Other methods include the use of a selective or specific protease capable of cleaving a CBD, e.g. a terminal CBD, from the remainder of the polypeptide chain of the protein in question.

As already indicated (*vide supra*), once a nucleotide sequence encoding the substrate-binding (carbohydrate-binding) region has been identified, either as cDNA or chromosomal DNA, it may then be manipulated in a variety of ways to fuse it to a DNA sequence encoding the enzyme or enzymatically active amino acid sequence of interest. The DNA fragment encoding the carbohydrate-binding amino acid sequence, and the DNA encoding the enzyme or enzymatically active amino acid sequence of interest are then ligated with or without a linker. The resulting ligated DNA may then be manipulated in a variety of ways to achieve expression. Preferred microbial expression hosts include certain *Aspergillus* species (e.g. *A. niger* or *A. oryzae*), *Bacillus* species, and organisms such as *Escherichia coli* or *Saccharomyces cerevisiae*.

#### The Cellulose Binding Domains of the present invention

The relative binding constant  $K_r$  for binding to regenerated cellulosic (amorphous - PASC phosphoric acid swollen cellulose) or crystalline cellulose (bacterial microcrystalline cellulose - BMCC) celluloses at 4°C and pH 7.0 is defined as  $K_r = [N_0]K_a$  and determined from the slope of a plot of  $1/[X_b]$  vs  $1/[X_f]$ , using a double-weighted, least-squares analysis; wherein  $[N_0]$  is the

concentration of binding sites in the absence of ligand ( $\text{mol}(\text{g of cellulose})^{-1}$ ),  $K_a$  is the equilibrium association constant ( $\text{L mol}^{-1}$ ),  $[X_b]$  is the concentration of bound ligand ( $\text{mol}(\text{g of cellulose})^{-1}$ ) and  $[X_f]$  is the concentration of free ligand (molar) (N.R. Gilkes et al., (1992) J. Biol. Chem. 267, pp. 6743-6749) such as described in P.E. Johnson et al., (1996) Biochem. 35, 13895-13906.

Microcrystalline cellulose (Avicel PH101) can be obtained from FMC International (Little Island, County Cork, Ireland). Bacterial microcrystalline cellulose (BMCC) can be prepared from cultures of *Acetobacter xylinum* (ATCC 23769) as described in Gilkes et al. 1992. Regenerated cellulose (PASC) can be obtained by phosphoric-acid treatment of Avicel PH101 as reported in J. B. Coutinho et al., (1992) Mol. Microbiol. 6, 1243-1252.

Suitable CBDs for the purpose of the present invention are the CBDs derived from a fungal or bacterial strain, preferably a bacterial strain, more preferably from Family II or IV according the current classification [See P. Tomme et al. (*op cit.*)]; having a relative binding constant ( $K_r$ -a) for binding to amorphous cellulose higher than 2.4l/gcellulose, preferably higher than 3.5l/gcellulose, more preferably higher than 4l/gcellulose cellulase.

More preferred CBDs are CBDs from family IV as currently classified, derived from organisms selected from *Cellulomonas fimi*, *Clostridium stecorarium* (NCIMB11754), *Clostridium cellulolyticum*, *Myxococcus xanthus*, *Streptomyces reticuli* and/or *Thermomonospora fusca*. Most preferred CBDs are the CBDs selected from CBDs of the tandem N-terminal cellulose binding domains from *Cellulomonas fimi*  $\beta$ -1,4-glucanase CenC which binds amorphous but not crystalline cellulose (P.E. Johnson et al., (1996) Biochem. 35, 13895-13906 and 14381-14394; J.B. Coutinho et al., (1992) Molecular Microbiology 6(9), pp. 1243-1252), CBD-dimer from *Clostridium stecorarium* (NCIMB11754) XynA and/or the CBD E3 from *Thermomonospora fusca*.

Preferably, the cellulose binding domains for the purpose of the present invention will be further characterised by having a relative binding constant for crystalline cellulose ( $K_r$ -c) lower than 1l/gcellulose, preferably lower than 0.5l/gcellulose.

More preferably, the modified enzyme of the present invention comprises a catalytically active amino acid sequence of a cellulolytic enzyme linked to an amino acid sequence comprising a Cellulose Binding Domain (CBD) further characterised by having a relative binding constant (Kr-a) for binding to amorphous cellulose to a relative binding constant for crystalline cellulose (Kr-c) ratio above 3.

Even more preferably for the purpose of the present invention, the modified enzyme comprising a catalytically active (enzymatically active) amino acid sequence of a cellulolytic enzyme linked to CBD as defined in the present invention, will provide an enzymatic hydrolysis activity on phosphoric acid-swollen cellulose higher than 60 moles glucose equivalents released per minute and per mole enzyme. Activities were determined at 37°C and pH7 (N. Din et al., (1995) Prog. Biotech. 10, pp. 261-270).

### Linking region

The term "linker" or "linking region" or "Middle region- MR" is intended to indicate a region that might adjoin the CBD and connect it to the catalytically active amino acid sequence of the cellulolytic enzyme. When present, this linking can be achieved chemically or by recombinant techniques.

An example of the recombinant technique describing the expression of an enzyme with the CBD of different origin is described in S. Karita et al., (1996) Journal of Fermentation and Bioengineering, Vol. 81, No. 6, pp. 553-556. Preferred linking regions are amino acid (peptides), some examples thereof are described in N.R. Gilkes et al., Microbiol. Rev. 55, 1991, pp. 303-315. The linking region can comprise from 1 to about 100 amino acid residues, in particular of from 2 to 40 amino acid residues, e.g. from 2 to 15 amino acid residues. As stated above, it is preferred to use amino acids which are less favored by the surrounding proteases. Suitable amino acid linking regions are the *Humicola insolens* family 45 cellulase linker, the NifA gene of *Klebsiella pneumoniae*-CiP linker, the *E. coli* OmpA gene-CiP linker, the E3 cellulase *Thermomonospora fusca* linker and the CenA cellulase linker.; preferably the



*Humicola insolens* family 45 cellulase linker and the E3 cellulase *Thermomonospora fusca* linker.

Non amino acid/proteinic compounds, referred to as "non-amino acid" can also be used for the linking of the catalytically active amino acid sequence to the CBD :

1) Suitable non-amino acid linking regions are the polyethylene glycol derivatives described in the Shearwater polymers, Inc. catalog of January 1996, such as the nucleophilic PEGs, the carboxyl PEGs, the electrophilically activated PEGs, the sulfhydryl-selective PEGs, the heterofunctional PEGs, the biotin PEGs, the vinyl derivatives, the PEG silanes and the PEG phospholipids. In particular, suitable non-amino acid linking regions are the heterofunctional PEG, (X-PEG-Y) polymers from Shearwater such as PEG(NPC)<sub>2</sub>, PEG-(NH<sub>2</sub>)<sub>2</sub>, t-BOC-NH-PEG-NH<sub>2</sub>, t-BOC-NH-PEG-CO<sub>2</sub>NHS, OH-PEG-NH-tBOC, Fmoc-NH-PEG-CO<sub>2</sub>NHS or PEG(NPC)<sub>2</sub> MW 3400 from Sigma, glutaric dialdehyde 50 wt% solution in water from Aldrich, disuccinimidyl suberate (DSS) from Sigma,  $\gamma$ -maleimidobutyric acid N-hydroxysuccinimide ester (GMBS) from Sigma, 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (EDC) from Sigma and dimethyl suberimidate hydrochloride (DMS) from Sigma.

2) Other suitable non-amino acid linking regions are 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide, N-ethyl-5-phenylisoaxolium-3-sulphonate, 1-cyclohexyl-3(2morpholinoethyl) carbodide metho-p-toluene sulphonate, N-ethoxycarbonyl-2-ethoxy 1,2, dihydroquinoline or glutaraldehyde.

3) Also suitable are the crosslinkers described in the 1999/2000 Pierce Products Catalogue from the Pierce Company, under the heading "Cross linking reagents : the SMPH, SMCC, LC-SMCC compounds, and preferably the Sulfo-KMUS compound.

Preferred chemical linking regions are PEG(NPC)<sub>2</sub>, (NH<sub>2</sub>)<sub>2</sub>-PEG, t-BOC-NH-PEG-NH<sub>2</sub>, MAL-PEG-NHS, VS-PEG-NHS polymers from Shearwater and/or the Sulfo-KMUS compound from Pierce.

### **Detergent components**

The laundry detergent and/or fabric care compositions of the invention will comprise at least one additional detergent and/or fabric care components. The

precise nature of these additional components, and levels of incorporation thereof will depend on the physical form of the composition, and the nature of the cleaning operation for which it is to be used.

The laundry detergent and/or fabric care compositions of the present invention preferably further comprise a detergent ingredient selected from cationic surfactants, dye transfer inhibiting polymers, builders - in particular zeolite A and sodium tripolyphosphate - and/or clays. These laundry detergent and/or fabric care compositions achieve improved fabric care, including improved anti-wrinkle, anti-bobbling and anti-shrinkage properties to fabrics, as well as provide enhanced static control, fabric softness, colour appearance and fabric anti-wear properties and benefits and improved fabric cleaning while preventing tensile strength loss.

The laundry detergent and/or fabric care compositions according to the invention can be liquid, paste, gels, bars, tablets, spray, foam, powder or granular forms. Granular compositions can also be in "compact" form, the liquid compositions can also be in a "concentrated" form.

The compositions of the invention may for example, be formulated as hand and machine laundry detergent compositions including laundry additive compositions and compositions suitable for use in the soaking and/or pretreatment of stained fabrics, rinse added fabric softener compositions. Pre-or post treatment of fabric include gel, spray and liquid fabric care compositions. A rinse cycle with or without the presence of softening agents is also contemplated.

When formulated as compositions suitable for use in a laundry machine washing method, the compositions of the invention preferably contain both a surfactant and a builder compound and additionally one or more detergent components preferably selected from organic polymeric compounds, bleaching agents, additional enzymes, suds suppressors, dispersants, lime-soap dispersants, soil suspension and anti-redeposition agents and corrosion inhibitors. Laundry compositions can also contain softening agents, as additional detergent components.

The compositions of the invention can also be used as detergent additive products in solid or liquid form. Such additive products are intended to supplement or boost the performance of conventional detergent compositions and can be added at any stage of the cleaning process.

If needed the density of the laundry detergent compositions herein ranges from 400 to 1200 g/litre, preferably 600 to 950 g/litre of composition measured at 20°C.

The "compact" form of the compositions herein is best reflected by density and, in terms of composition, by the amount of inorganic filler salt; inorganic filler salts are conventional ingredients of detergent compositions in powder form; in conventional detergent compositions, the filler salts are present in substantial amounts, typically 17-35% by weight of the total composition. In the compact compositions, the filler salt is present in amounts not exceeding 15% of the total composition, preferably not exceeding 10%, most preferably not exceeding 5% by weight of the composition. The inorganic filler salts, such as meant in the present compositions are selected from the alkali and alkaline-earth-metal salts of sulphates and chlorides. A preferred filler salt is sodium sulphate.

Liquid detergent compositions according to the present invention can also be in a "concentrated form", in such case, the liquid detergent compositions according the present invention will contain a lower amount of water, compared to conventional liquid detergents. Typically the water content of the concentrated liquid detergent is preferably less than 40%, more preferably less than 30%, most preferably less than 20% by weight of the detergent composition.

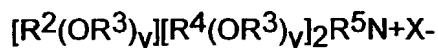
### **Surfactant system**

The laundry detergent and/or fabric care compositions according to the present invention comprise a surfactant system wherein the surfactant can be selected from nonionic and/or anionic and/or cationic and/or ampholytic and/or zwitterionic and/or semi-polar surfactants. Preferred surfactants are cationic surfactants. It has been surprisingly found that the laundry detergent and/or fabric care compositions further comprising a cationic surfactant, achieve improved fabric care, including improved anti-wrinkle, anti-bobbling and anti-shrinkage properties to fabrics, as well as provide enhanced static control, fabric softness, colour appearance and fabric anti-wear properties and benefits and improved fabric cleaning while preventing tensile strength loss.

The surfactant is typically present at a level of from 0.1% to 60% by weight. More preferred levels of incorporation are 1% to 35% by weight, most preferably from 1% to 30% by weight of laundry detergent and/or fabric care compositions in accord with the invention.

The surfactant is preferably formulated to be compatible with enzyme components present in the composition. In liquid or gel compositions the surfactant is most preferably formulated such that it promotes, or at least does not degrade, the stability of any enzyme in these compositions.

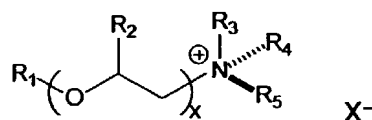
Cationic deterative surfactants suitable for use in the laundry detergent and/or fabric care compositions of the present invention are those having one long-chain hydrocarbyl group. Examples of such cationic surfactants include the ammonium surfactants such as alkyltrimethylammonium halogenides, and those surfactants having the formula :



wherein  $R^2$  is an alkyl or alkyl benzyl group having from about 8 to about 18 carbon atoms in the alkyl chain, each  $R^3$  is selected from the group consisting of  $-CH_2CH_2-$ ,  $-CH_2CH(CH_3)-$ ,  $-CH_2CH(CH_2OH)-$ ,  $-CH_2CH_2CH_2-$ , and mixtures thereof; each  $R^4$  is selected from the group consisting of  $C_1-C_4$  alkyl,  $C_1-C_4$  hydroxyalkyl, benzyl ring structures formed by joining the two  $R^4$  groups, -

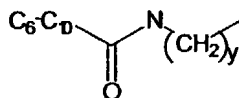
$\text{CH}_2\text{CHOH-CHOHCOR}^6\text{CHOHCH}_2\text{OH}$  wherein  $\text{R}^6$  is any hexose or hexose polymer having a molecular weight less than about 1000, and hydrogen when  $y$  is not 0;  $\text{R}^5$  is the same as  $\text{R}^4$  or is an alkyl chain wherein the total number of carbon atoms of  $\text{R}^2$  plus  $\text{R}^5$  is not more than about 18; each  $y$  is from 0 to about 10 and the sum of the  $y$  values is from 0 to about 15; and  $\text{X}$  is any compatible anion.

Quaternary ammonium surfactant suitable for the present invention has the formula (I):



Formula I

whereby  $\text{R}^1$  is a short chainlength alkyl (C6-C10) or alkylamidoalkyl of the formula (II):



Formula II

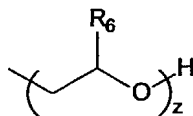
$y$  is 2-4, preferably 3.

whereby  $\text{R}^2$  is H or a C1-C3 alkyl,

whereby  $x$  is 0-4, preferably 0-2, most preferably 0,

whereby  $\text{R}^3$ ,  $\text{R}^4$  and  $\text{R}^5$  are either the same or different and can be either a short chain alkyl (C1-C3) or alkoxyated alkyl of the formula III,

whereby  $\text{X}^-$  is a counterion, preferably a halide, e.g. chloride or methylsulfate.



Formula III

R6 is C<sub>1</sub>-C<sub>4</sub> and z is 1 or 2.

Preferred quat ammonium surfactants are those as defined in formula I whereby R<sub>1</sub> is C<sub>8</sub>, C<sub>10</sub> or mixtures thereof, x=0, R<sub>3</sub>, R<sub>4</sub> = CH<sub>3</sub> and R<sub>5</sub> = CH<sub>2</sub>CH<sub>2</sub>OH.

Highly preferred cationic surfactants are the water-soluble quaternary ammonium compounds useful in the present composition having the formula :



wherein R<sub>1</sub> is C<sub>8</sub>-C<sub>16</sub> alkyl, each of R<sub>2</sub>, R<sub>3</sub> and R<sub>4</sub> is independently C<sub>1</sub>-C<sub>4</sub> alkyl, C<sub>1</sub>-C<sub>4</sub> hydroxy alkyl, benzyl, and -(C<sub>2</sub>H<sub>4</sub>O)<sub>x</sub>H where x has a value from 2 to 5, and X is an anion. Not more than one of R<sub>2</sub>, R<sub>3</sub> or R<sub>4</sub> should be benzyl.

The preferred alkyl chain length for R<sub>1</sub> is C<sub>12</sub>-C<sub>15</sub> particularly where the alkyl group is a mixture of chain lengths derived from coconut or palm kernel fat or is derived synthetically by olefin build up or OXO alcohols synthesis. Preferred groups for R<sub>2</sub>R<sub>3</sub> and R<sub>4</sub> are methyl and hydroxyethyl groups and the anion X may be selected from halide, methosulphate, acetate and phosphate ions.

Examples of suitable quaternary ammonium compounds of formulae (i) for use herein are :

- coconut trimethyl ammonium chloride or bromide;
- coconut methyl dihydroxyethyl ammonium chloride or bromide;
- decyl triethyl ammonium chloride;
- decyl dimethyl hydroxyethyl ammonium chloride or bromide;
- C<sub>12</sub>-15 dimethyl hydroxyethyl ammonium chloride or bromide;
- coconut dimethyl hydroxyethyl ammonium chloride or bromide;
- myristyl trimethyl ammonium methyl sulphate;
- lauryl dimethyl benzyl ammonium chloride or bromide;
- lauryl dimethyl (ethenoxy)<sub>4</sub> ammonium chloride or bromide;
- choline esters (compounds of formula (i) wherein R<sub>1</sub> is CH<sub>2</sub>-CH<sub>2</sub>-O-C-C<sub>12-14</sub> alkyl and R<sub>2</sub>R<sub>3</sub>R<sub>4</sub> are methyl).



di-alkyl imidazolines [compounds of formula (i)].

Other cationic surfactants useful herein are also described in U.S. Patent 4,228,044, Cambre, issued October 14, 1980 and in European Patent Application EP 000,224.

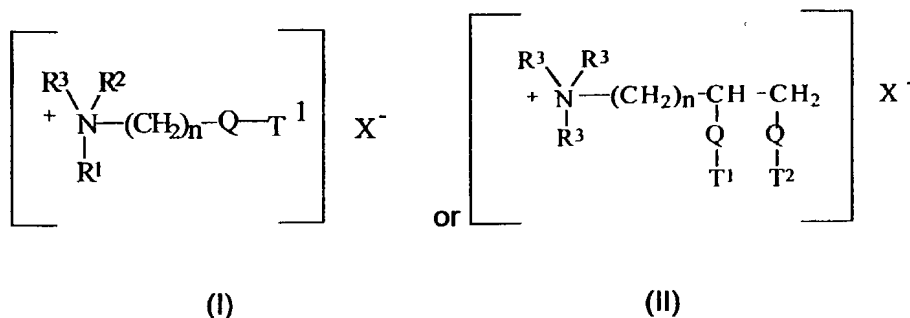
Typical cationic fabric softening components include the water-insoluble quaternary-ammonium fabric softening actives or the corresponding amine precursor, the most commonly used having been di-long alkyl chain ammonium chloride or methyl sulfate.

Preferred cationic softeners among these include the following:

- 1) ditallow dimethylammonium chloride (DTDMAC);
- 2) dihydrogenated tallow dimethylammonium chloride;
- 3) dihydrogenated tallow dimethylammonium methylsulfate;
- 4) distearyl dimethylammonium chloride;
- 5) dioleyl dimethylammonium chloride;
- 6) dipalmityl hydroxyethyl methylammonium chloride;
- 7) stearyl benzyl dimethylammonium chloride;
- 8) tallow trimethylammonium chloride;
- 9) hydrogenated tallow trimethylammonium chloride;
- 10) C<sub>12-14</sub> alkyl hydroxyethyl dimethylammonium chloride;
- 11) C<sub>12-18</sub> alkyl dihydroxyethyl methylammonium chloride;
- 12) di(stearoyloxyethyl) dimethylammonium chloride (DSOEDMAC);
- 13) di(tallow-oxy-ethyl) dimethylammonium chloride;
- 14) ditallow imidazolinium methylsulfate;
- 15) 1-(2-tallowylamidoethyl)-2-tallowyl imidazolinium methylsulfate.

Biodegradable quaternary ammonium compounds have been presented as alternatives to the traditionally used di-long alkyl chain ammonium chlorides and methyl sulfates. Such quaternary ammonium compounds contain long chain alk(en)yl groups interrupted by functional groups such as carboxy groups. Said materials and fabric softening compositions containing them are disclosed in numerous publications such as EP-A-0,040,562, and EP-A-0,239,910.

The quaternary ammonium compounds and amine precursors herein have the formula (I) or (II), below :



wherein Q is selected from -O-C(O)-, -C(O)-O-, -O-C(O)-O-, -NR<sup>4</sup>-C(O)-, -C(O)-NR<sup>4</sup>-;

R<sup>1</sup> is (CH<sub>2</sub>)<sub>n</sub>-Q-T<sup>2</sup> or T<sup>3</sup>;

R<sup>2</sup> is (CH<sub>2</sub>)<sub>m</sub>-Q-T<sup>4</sup> or T<sup>5</sup> or R<sup>3</sup>;

R<sup>3</sup> is C<sub>1</sub>-C<sub>4</sub> alkyl or C<sub>1</sub>-C<sub>4</sub> hydroxyalkyl or H;

R<sup>4</sup> is H or C<sub>1</sub>-C<sub>4</sub> alkyl or C<sub>1</sub>-C<sub>4</sub> hydroxyalkyl;

T<sup>1</sup>, T<sup>2</sup>, T<sup>3</sup>, T<sup>4</sup>, T<sup>5</sup> are independently C<sub>11</sub>-C<sub>22</sub> alkyl or alkenyl;

n and m are integers from 1 to 4; and

X<sup>-</sup> is a softener-compatible anion.

Non-limiting examples of softener-compatible anions include chloride or methyl sulfate.

The alkyl, or alkenyl, chain T<sup>1</sup>, T<sup>2</sup>, T<sup>3</sup>, T<sup>4</sup>, T<sup>5</sup> must contain at least 11 carbon atoms, preferably at least 16 carbon atoms. The chain may be straight or branched.

Tallow is a convenient and inexpensive source of long chain alkyl and alkenyl material. The compounds wherein T<sup>1</sup>, T<sup>2</sup>, T<sup>3</sup>, T<sup>4</sup>, T<sup>5</sup> represents the mixture of long chain materials typical for tallow are particularly preferred.

Specific examples of quaternary ammonium compounds suitable for use in the aqueous fabric softening compositions herein include :

- 1) N,N-di(tallowyl-oxy-ethyl)-N,N-dimethyl ammonium chloride;
- 2) N,N-di(tallowyl-oxy-ethyl)-N-methyl, N-(2-hydroxyethyl) ammonium methyl sulfate;
- 3) N,N-di(2-tallowyl-oxy-2-oxo-ethyl)-N,N-dimethyl ammonium chloride;



- 4) N,N-di(2-tallowyl-oxy-ethylcarbonyl-oxy-ethyl)-N,N-dimethyl ammonium chloride;
- 5) N-(2-tallowyl-oxy-2-ethyl)-N-(2-tallowyl-oxy-2-oxo-ethyl)-N,N-dimethyl ammonium chloride;
- 6) N,N,N-tri(tallowyl-oxy-ethyl)-N-methyl ammonium chloride;
- 7) N-(2-tallowyl-oxy-2-oxo-ethyl)-N-(tallowyl-N,N-dimethyl-ammonium chloride; and
- 8) 1,2-ditallowyl-oxy-3-trimethylammonio propane chloride; and mixtures of any of the above materials.

When included therein, the laundry detergent and/or fabric care compositions of the present invention typically comprise from 0.2% to about 25%, preferably from about 1% to about 8% by weight of such cationic surfactants.

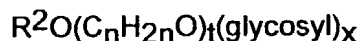
Polyethylene, polypropylene, and polybutylene oxide condensates of alkyl phenols are suitable for use as the nonionic surfactant of the surfactant systems of the present invention, with the polyethylene oxide condensates being preferred. These compounds include the condensation products of alkyl phenols having an alkyl group containing from about 6 to about 14 carbon atoms, preferably from about 8 to about 14 carbon atoms, in either a straight-chain or branched-chain configuration with the alkylene oxide. In a preferred embodiment, the ethylene oxide is present in an amount equal to from about 2 to about 25 moles, more preferably from about 3 to about 15 moles, of ethylene oxide per mole of alkyl phenol. Commercially available nonionic surfactants of this type include Igepal<sup>TM</sup> CO-630, marketed by the GAF Corporation; and Triton<sup>TM</sup> X-45, X-114, X-100 and X-102, all marketed by the Rohm & Haas Company. These surfactants are commonly referred to as alkylphenol alkoxyates (e.g., alkyl phenol ethoxylates).

The condensation products of primary and secondary aliphatic alcohols with from about 1 to about 25 moles of ethylene oxide are suitable for use as the nonionic surfactant of the nonionic surfactant systems of the present invention. The alkyl chain of the aliphatic alcohol can either be straight or branched, primary or secondary, and generally contains from about 8 to about 22 carbon atoms. Preferred are the condensation products of alcohols having an alkyl

group containing from about 8 to about 20 carbon atoms, more preferably from about 10 to about 18 carbon atoms, with from about 2 to about 10 moles of ethylene oxide per mole of alcohol. About 2 to about 7 moles of ethylene oxide and most preferably from 2 to 5 moles of ethylene oxide per mole of alcohol are present in said condensation products. Examples of commercially available nonionic surfactants of this type include Tergitol<sup>TM</sup> 15-S-9 (the condensation product of C<sub>11</sub>-C<sub>15</sub> linear alcohol with 9 moles ethylene oxide), Tergitol<sup>TM</sup> 24-L-6 NMW (the condensation product of C<sub>12</sub>-C<sub>14</sub> primary alcohol with 6 moles ethylene oxide with a narrow molecular weight distribution), both marketed by Union Carbide Corporation; Neodol<sup>TM</sup> 45-9 (the condensation product of C<sub>14</sub>-C<sub>15</sub> linear alcohol with 9 moles of ethylene oxide), Neodol<sup>TM</sup> 23-3 (the condensation product of C<sub>12</sub>-C<sub>13</sub> linear alcohol with 3.0 moles of ethylene oxide), Neodol<sup>TM</sup> 45-7 (the condensation product of C<sub>14</sub>-C<sub>15</sub> linear alcohol with 7 moles of ethylene oxide), Neodol<sup>TM</sup> 45-5 (the condensation product of C<sub>14</sub>-C<sub>15</sub> linear alcohol with 5 moles of ethylene oxide) marketed by Shell Chemical Company, Kyro<sup>TM</sup> EOB (the condensation product of C<sub>13</sub>-C<sub>15</sub> alcohol with 9 moles ethylene oxide), marketed by The Procter & Gamble Company, and Genapol LA O3O or O5O (the condensation product of C<sub>12</sub>-C<sub>14</sub> alcohol with 3 or 5 moles of ethylene oxide) marketed by Hoechst. Preferred range of HLB in these products is from 8-11 and most preferred from 8-10.

Also useful as the nonionic surfactant of the surfactant systems of the present invention are the alkylpolysaccharides disclosed in U.S. Patent 4,565,647, Llenado, issued January 21, 1986, having a hydrophobic group containing from about 6 to about 30 carbon atoms, preferably from about 10 to about 16 carbon atoms and a polysaccharide, e.g. a polyglycoside, hydrophilic group containing from about 1.3 to about 10, preferably from about 1.3 to about 3, most preferably from about 1.3 to about 2.7 saccharide units. Any reducing saccharide containing 5 or 6 carbon atoms can be used, e.g., glucose, galactose and galactosyl moieties can be substituted for the glucosyl moieties (optionally the hydrophobic group is attached at the 2-, 3-, 4-, etc. positions thus giving a glucose or galactose as opposed to a glucoside or galactoside). The intersaccharide bonds can be, e.g., between the one position of the additional saccharide units and the 2-, 3-, 4-, and/or 6- positions on the preceding saccharide units.

The preferred alkylpolyglycosides have the formula



wherein  $R^2$  is selected from the group consisting of alkyl, alkylphenyl, hydroxyalkyl, hydroxyalkylphenyl, and mixtures thereof in which the alkyl groups contain from about 10 to about 18, preferably from about 12 to about 14, carbon atoms;  $n$  is 2 or 3, preferably 2;  $t$  is from 0 to about 10, preferably 0; and  $x$  is from about 1.3 to about 10, preferably from about 1.3 to about 3, most preferably from about 1.3 to about 2.7. The glycosyl is preferably derived from glucose. To prepare these compounds, the alcohol or alkylpolyethoxy alcohol is formed first and then reacted with glucose, or a source of glucose, to form the glucoside (attachment at the 1-position). The additional glycosyl units can then be attached between their 1-position and the preceding glycosyl units 2-, 3-, 4- and/or 6-position, preferably predominately the 2-position.

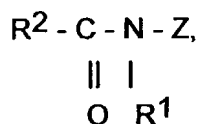
The condensation products of ethylene oxide with a hydrophobic base formed by the condensation of propylene oxide with propylene glycol are also suitable for use as the additional nonionic surfactant systems of the present invention. The hydrophobic portion of these compounds will preferably have a molecular weight of from about 1500 to about 1800 and will exhibit water insolubility. The addition of polyoxyethylene moieties to this hydrophobic portion tends to increase the water solubility of the molecule as a whole, and the liquid character of the product is retained up to the point where the polyoxyethylene content is about 50% of the total weight of the condensation product, which corresponds to condensation with up to about 40 moles of ethylene oxide. Examples of compounds of this type include certain of the commercially-available Plurafac<sup>TM</sup> LF404 and Pluronic<sup>TM</sup> surfactants, marketed by BASF.

Also suitable for use as the nonionic surfactant of the nonionic surfactant system of the present invention, are the condensation products of ethylene oxide with the product resulting from the reaction of propylene oxide and ethylenediamine. The hydrophobic moiety of these products consists of the reaction product of ethylenediamine and excess propylene oxide, and generally has a molecular weight of from about 2500 to about 3000. This hydrophobic moiety is condensed with ethylene oxide to the extent that the condensation product

contains from about 40% to about 80% by weight of polyoxyethylene and has a molecular weight of from about 5,000 to about 11,000. Examples of this type of nonionic surfactant include certain of the commercially available Tetronic<sup>TM</sup> compounds, marketed by BASF.

Preferred for use as the nonionic surfactant of the surfactant systems of the present invention are polyethylene oxide condensates of alkyl phenols, condensation products of primary and secondary aliphatic alcohols with from about 1 to about 25 moles of ethylene oxide, alkylpolysaccharides, and mixtures thereof. Most preferred are C<sub>8</sub>-C<sub>14</sub> alkyl phenol ethoxylates having from 3 to 15 ethoxy groups and C<sub>8</sub>-C<sub>18</sub> alcohol ethoxylates (preferably C<sub>10</sub> avg.) having from 2 to 10 ethoxy groups, and mixtures thereof.

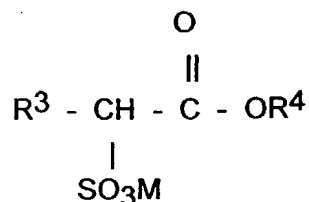
Highly preferred nonionic surfactants are polyhydroxy fatty acid amide surfactants of the formula.



wherein R<sup>1</sup> is H, or R<sup>1</sup> is C<sub>1-4</sub> hydrocarbyl, 2-hydroxy ethyl, 2-hydroxy propyl or a mixture thereof, R<sup>2</sup> is C<sub>5-31</sub> hydrocarbyl, and Z is a polyhydroxyhydrocarbyl having a linear hydrocarbyl chain with at least 3 hydroxyls directly connected to the chain, or an alkoxylated derivative thereof. Preferably, R<sup>1</sup> is methyl, R<sup>2</sup> is a straight C<sub>11-15</sub> alkyl or C<sub>16-18</sub> alkyl or alkenyl chain such as coconut alkyl or mixtures thereof, and Z is derived from a reducing sugar such as glucose, fructose, maltose, lactose, in a reductive amination reaction.

Suitable anionic surfactants to be used are linear alkyl benzene sulfonate, alkyl ester sulfonate surfactants including linear esters of C<sub>8</sub>-C<sub>20</sub> carboxylic acids (i.e., fatty acids) which are sulfonated with gaseous SO<sub>3</sub> according to "The Journal of the American Oil Chemists Society", 52 (1975), pp. 323-329. Suitable starting materials would include natural fatty substances as derived from tallow, palm oil, etc.

The preferred alkyl ester sulfonate surfactant, especially for laundry applications, comprise alkyl ester sulfonate surfactants of the structural formula:



wherein R<sup>3</sup> is a C<sub>8</sub>-C<sub>20</sub> hydrocarbyl, preferably an alkyl, or combination thereof, R<sup>4</sup> is a C<sub>1</sub>-C<sub>6</sub> hydrocarbyl, preferably an alkyl, or combination thereof, and M is a cation which forms a water soluble salt with the alkyl ester sulfonate. Suitable salt-forming cations include metals such as sodium, potassium, and lithium, and substituted or unsubstituted ammonium cations, such as mono-ethanolamine, diethanolamine, and triethanolamine. Preferably, R<sup>3</sup> is C<sub>10</sub>-C<sub>16</sub> alkyl, and R<sup>4</sup> is methyl, ethyl or isopropyl. Especially preferred are the methyl ester sulfonates wherein R<sup>3</sup> is C<sub>10</sub>-C<sub>16</sub> alkyl.

Other suitable anionic surfactants include the alkyl sulfate surfactants which are water soluble salts or acids of the formula ROSO<sub>3</sub>M wherein R preferably is a C<sub>10</sub>-C<sub>24</sub> hydrocarbyl, preferably an alkyl or hydroxyalkyl having a C<sub>10</sub>-C<sub>20</sub> alkyl component, more preferably a C<sub>12</sub>-C<sub>18</sub> alkyl or hydroxyalkyl, and M is H or a cation, e.g., an alkali metal cation (e.g. sodium, potassium, lithium), or ammonium or substituted ammonium (e.g. methyl-, dimethyl-, and trimethyl ammonium cations and quaternary ammonium cations such as tetramethylammonium and dimethyl piperdinium cations and quaternary ammonium cations derived from alkylamines such as ethylamine, diethylamine, triethylamine, and mixtures thereof, and the like). Typically, alkyl chains of C<sub>12</sub>-C<sub>16</sub> are preferred for lower wash temperatures (e.g. below about 50°C) and C<sub>16</sub>-18 alkyl chains are preferred for higher wash temperatures (e.g. above about 50°C).

Other anionic surfactants useful for deterative purposes can also be included in the laundry detergent and/or fabric care compositions of the present invention. These can include salts (including, for example, sodium, potassium, ammonium, and substituted ammonium salts such as mono-, di- and triethanolamine salts) of soap, C<sub>8</sub>-C<sub>22</sub> primary or secondary alkanesulfonates, C<sub>8</sub>-C<sub>24</sub> olefinsulfonates, sulfonated polycarboxylic acids prepared by sulfonation of the pyrolyzed product of alkaline earth metal citrates, e.g., as described in British patent specification No. 1,082,179, C<sub>8</sub>-C<sub>24</sub> alkylpolyglycoethersulfates (containing up to 10 moles of ethylene oxide); alkyl glycerol sulfonates, fatty acyl glycerol sulfonates, fatty

oleyl glycerol sulfates, alkyl phenol ethylene oxide ether sulfates, paraffin sulfonates, alkyl phosphates, isethionates such as the acyl isethionates, N-acyl taurates, alkyl succinamates and sulfosuccinates, monoesters of sulfosuccinates (especially saturated and unsaturated C<sub>12</sub>-C<sub>18</sub> monoesters) and diesters of sulfosuccinates (especially saturated and unsaturated C<sub>6</sub>-C<sub>12</sub> diesters), acyl sarcosinates, sulfates of alkylpolysaccharides such as the sulfates of alkylpolyglucoside (the nonionic nonsulfated compounds being described below), branched primary alkyl sulfates, and alkyl polyethoxy carboxylates such as those of the formula  $RO(CH_2CH_2O)_k-CH_2COO-M^+$  wherein R is a C<sub>8</sub>-C<sub>22</sub> alkyl, k is an integer from 1 to 10, and M is a soluble salt-forming cation. Resin acids and hydrogenated resin acids are also suitable, such as rosin, hydrogenated rosin, and resin acids and hydrogenated resin acids present in or derived from tall oil.

Further examples are described in "Surface Active Agents and Detergents" (Vol. I and II by Schwartz, Perry and Berch). A variety of such surfactants are also generally disclosed in U.S. Patent 3,929,678, issued December 30, 1975 to Laughlin, et al. at Column 23, line 58 through Column 29, line 23 (herein incorporated by reference).

When included therein, the laundry detergent compositions of the present invention typically comprise from about 1% to about 40%, preferably from about 3% to about 20% by weight of such anionic surfactants.

Highly preferred anionic surfactants include alkyl alkoxyated sulfate surfactants hereof are water soluble salts or acids of the formula  $RO(A)_mSO_3M$  wherein R is an unsubstituted C<sub>10</sub>-C<sub>24</sub> alkyl or hydroxyalkyl group having a C<sub>10</sub>-C<sub>24</sub> alkyl component, preferably a C<sub>12</sub>-C<sub>20</sub> alkyl or hydroxyalkyl, more preferably C<sub>12</sub>-C<sub>18</sub> alkyl or hydroxyalkyl, A is an ethoxy or propoxy unit, m is greater than zero, typically between about 0.5 and about 6, more preferably between about 0.5 and about 3, and M is H or a cation which can be, for example, a metal cation (e.g., sodium, potassium, lithium, calcium, magnesium, etc.), ammonium or substituted-ammonium cation. Alkyl ethoxyated sulfates as well as alkyl propoxyated sulfates are contemplated herein. Specific examples of substituted ammonium cations include methyl-, dimethyl-, trimethyl-ammonium cations and quaternary ammonium cations such as tetramethyl-ammonium and dimethyl piperdinium cations and those derived from alkylamines such as ethylamine,

diethylamine, triethylamine, mixtures thereof, and the like. Exemplary surfactants are C<sub>12</sub>-C<sub>18</sub> alkyl polyethoxylate (1.0) sulfate (C<sub>12</sub>-C<sub>18</sub>E(1.0)M), C<sub>12</sub>-C<sub>18</sub> alkyl polyethoxylate (2.25) sulfate (C<sub>12</sub>-C<sub>18</sub>E(2.25)M), C<sub>12</sub>-C<sub>18</sub> alkyl polyethoxylate (3.0) sulfate (C<sub>12</sub>-C<sub>18</sub>E(3.0)M), and C<sub>12</sub>-C<sub>18</sub> alkyl polyethoxylate (4.0) sulfate (C<sub>12</sub>-C<sub>18</sub>E(4.0)M), wherein M is conveniently selected from sodium and potassium.

The laundry detergent and/or fabric care compositions of the present invention may also contain ampholytic, zwitterionic, and semi-polar surfactants, as well as the nonionic and/or anionic surfactants other than those already described herein.

Ampholytic surfactants are also suitable for use in the laundry detergent and/or fabric care compositions of the present invention. These surfactants can be broadly described as aliphatic derivatives of secondary or tertiary amines, or aliphatic derivatives of heterocyclic secondary and tertiary amines in which the aliphatic radical can be straight- or branched-chain. One of the aliphatic substituents contains at least about 8 carbon atoms, typically from about 8 to about 18 carbon atoms, and at least one contains an anionic water-solubilizing group, e.g. carboxy, sulfonate, sulfate. See U.S. Patent No. 3,929,678 to Laughlin et al., issued December 30, 1975 at column 19, lines 18-35, for examples of ampholytic surfactants.

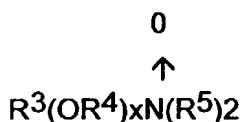
When included therein, the laundry detergent and/or fabric care compositions of the present invention typically comprise from 0.2% to about 15%, preferably from about 1% to about 10% by weight of such ampholytic surfactants.

Zwitterionic surfactants are also suitable for use in laundry detergent and/or fabric care compositions. These surfactants can be broadly described as derivatives of secondary and tertiary amines, derivatives of heterocyclic secondary and tertiary amines, or derivatives of quaternary ammonium, quaternary phosphonium or tertiary sulfonium compounds. See U.S. Patent No. 3,929,678 to Laughlin et al., issued December 30, 1975 at column 19, line 38 through column 22, line 48, for examples of zwitterionic surfactants.

When included therein, the laundry detergent and/or fabric care compositions of the present invention typically comprise from 0.2% to about 15%, preferably from about 1% to about 10% by weight of such zwitterionic surfactants.

Semi-polar nonionic surfactants are a special category of nonionic surfactants which include water-soluble amine oxides containing one alkyl moiety of from about 10 to about 18 carbon atoms and 2 moieties selected from the group consisting of alkyl groups and hydroxyalkyl groups containing from about 1 to about 3 carbon atoms; water-soluble phosphine oxides containing one alkyl moiety of from about 10 to about 18 carbon atoms and 2 moieties selected from the group consisting of alkyl groups and hydroxyalkyl groups containing from about 1 to about 3 carbon atoms; and water-soluble sulfoxides containing one alkyl moiety of from about 10 to about 18 carbon atoms and a moiety selected from the group consisting of alkyl and hydroxyalkyl moieties of from about 1 to about 3 carbon atoms.

Semi-polar nonionic detergent surfactants include the amine oxide surfactants having the formula



wherein  $R^3$  is an alkyl, hydroxyalkyl, or alkyl phenyl group or mixtures thereof containing from about 8 to about 22 carbon atoms;  $R^4$  is an alkylene or hydroxyalkylene group containing from about 2 to about 3 carbon atoms or mixtures thereof;  $x$  is from 0 to about 3; and each  $R^5$  is an alkyl or hydroxyalkyl group containing from about 1 to about 3 carbon atoms or a polyethylene oxide group containing from about 1 to about 3 ethylene oxide groups. The  $R^5$  groups can be attached to each other, e.g., through an oxygen or nitrogen atom, to form a ring structure.

These amine oxide surfactants in particular include  $C_{10}$ - $C_{18}$  alkyl dimethyl amine oxides and  $C_8$ - $C_{12}$  alkoxy ethyl dihydroxy ethyl amine oxides.

When included therein, the cleaning compositions of the present invention typically comprise from 0.2% to about 15%, preferably from about 1% to about 10% by weight of such semi-polar nonionic surfactants.

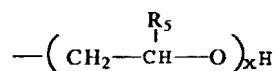


The laundry detergent and/or fabric care composition of the present invention may further comprise a cosurfactant selected from the group of primary or tertiary amines.

Suitable primary amines for use herein include amines according to the formula  $R_1NH_2$  wherein  $R_1$  is a  $C_6$ - $C_{12}$ , preferably  $C_6$ - $C_{10}$  alkyl chain or  $R_4X(CH_2)_n$ ,  $X$  is  $-O-$ ,  $-C(O)NH-$  or  $-NH-$ ,  $R_4$  is a  $C_6$ - $C_{12}$  alkyl chain  $n$  is between 1 to 5, preferably 3.  $R_1$  alkyl chains may be straight or branched and may be interrupted with up to 12, preferably less than 5 ethylene oxide moieties.

Preferred amines according to the formula herein above are  $n$ -alkyl amines. Suitable amines for use herein may be selected from 1-hexylamine, 1-octylamine, 1-decylamine and laurylamine. Other preferred primary amines include  $C_8$ - $C_{10}$  oxypropylamine, octyloxypropylamine, 2-ethylhexyloxypropylamine, lauryl amido propylamine and amido propylamine.

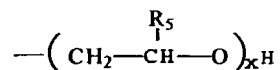
Suitable tertiary amines for use herein include tertiary amines having the formula  $R_1R_2R_3N$  wherein  $R_1$  and  $R_2$  are  $C_1$ - $C_8$  alkyl chains or



$R_3$  is either a  $C_6$ - $C_{12}$ , preferably  $C_6$ - $C_{10}$  alkyl chain, or  $R_3$  is  $R_4X(CH_2)_n$ , whereby  $X$  is  $-O-$ ,  $-C(O)NH-$  or  $-NH-$ ,  $R_4$  is a  $C_4$ - $C_{12}$ ,  $n$  is between 1 to 5, preferably 2-3.  $R_5$  is H or  $C_1$ - $C_2$  alkyl and  $x$  is between 1 to 6.

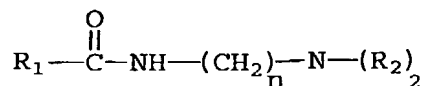
$R_3$  and  $R_4$  may be linear or branched;  $R_3$  alkyl chains may be interrupted with up to 12, preferably less than 5, ethylene oxide moieties.

Preferred tertiary amines are  $R_1R_2R_3N$  where  $R_1$  is a  $C_6$ - $C_{12}$  alkyl chain,  $R_2$  and  $R_3$  are  $C_1$ - $C_3$  alkyl or



where  $R_5$  is H or  $CH_3$  and  $x = 1-2$ .

Also preferred are the amidoamines of the formula:



wherein  $R_1$  is  $C_6$ - $C_{12}$  alkyl;  $n$  is 2-4,  
preferably  $n$  is 3;  $R_2$  and  $R_3$  is  $C_1$ - $C_4$

Most preferred amines of the present invention include 1-octylamine, 1-hexylamine, 1-decylamine, 1-dodecylamine, C8-10oxypropylamine, N coco 1-3diaminopropane, coconutalkyldimethylamine, lauryldimethylamine, lauryl bis(hydroxyethyl)amine, coco bis(hydroxyethyl)amine, lauryl amine 2 moles propoxylated, octyl amine 2 moles propoxylated, lauryl amidopropyl-dimethylamine, C8-10 amidopropyl-dimethylamine and C10 amidopropyl-dimethylamine.

The most preferred amines for use in the compositions herein are 1-hexylamine, 1-octylamine, 1-decylamine, 1-dodecylamine. Especially desirable are  $n$ -dodecyl-dimethylamine and bis(hydroxyethyl)coconutalkylamine and oleylamine 7 times ethoxylated, lauryl amido propylamine and cocoamido propylamine.

### ***Conventional detergent enzymes***

The laundry detergent and/or fabric care compositions can in addition to the modified cellulase, further comprise one or more enzymes which provide cleaning performance and/or fabric care benefits.

Said other enzymes include enzymes selected from cellulases without naturally occurring CBDs or with a having a relative binding constant ( $K_r$ -a) for binding to amorphous cellulose lower than 2.4l/gcellulose, hemicellulases, peroxidases, proteases, gluco-amylases, amylases, xylanases, lipases, phospholipases, esterases, cutinases, pectinases, keratanases, reductases, oxidases, phenoloxidases, lipoxygenases, ligninases, pullulanases, tannases, pentosanases, malanases,  $\beta$ -glucanases, arabinosidases, hyaluronidase, chondroitinase, laccase or mixtures thereof.

A preferred combination is a laundry detergent and/or fabric care composition having cocktail of conventional applicable enzymes like protease, amylase, lipase, cutinase and/or cellulase in conjunction with one or more plant cell wall degrading enzymes.

Suitable proteases are the subtilisins which are obtained from particular strains of *B. subtilis* and *B. licheniformis* (subtilisin BPN and BPN'). One suitable protease is obtained from a strain of *Bacillus*, having maximum activity throughout the pH range of 8-12, developed and sold as ESPERASE® by Novo Industries A/S of Denmark, hereinafter "Novo". The preparation of this enzyme and analogous enzymes is described in GB 1,243,784 to Novo. Other suitable proteases include ALCALASE®, DURAZYM® and SAVINASE® from Novo and MAXATASE®, MAXACAL®, PROPERASE® and MAXAPEM® (protein engineered Maxacal) from Gist-Brocades. Proteolytic enzymes also encompass modified bacterial serine proteases, such as those described in European Patent Application Serial Number 87 303761.8, filed April 28, 1987 (particularly pages 17, 24 and 98), and which is called herein "Protease B", and in European Patent Application 199,404, Venegas, published October 29, 1986, which refers to a modified bacterial serine proteolytic enzyme which is called "Protease A" herein. Suitable is what is called herein "Protease C", which is a variant of an alkaline serine protease from *Bacillus* in which lysine replaced arginine at position 27, tyrosine replaced valine at position 104, serine replaced asparagine at position 123, and alanine replaced threonine at position 274. Protease C is described in EP 90915958:4, corresponding to WO 91/06637, Published May 16, 1991. Genetically modified variants, particularly of Protease C, are also included herein.

A preferred protease referred to as "Protease D" is a carbonyl hydrolase variant having an amino acid sequence not found in nature, which is derived from a precursor carbonyl hydrolase by substituting a different amino acid for a plurality of amino acid residues at a position in said carbonyl hydrolase equivalent to position +76, preferably also in combination with one or more amino acid residue positions equivalent to those selected from the group consisting of +99, +101, +103, +104, +107, +123, +27, +105, +109, +126, +128, +135, +156, +166, +195, +197, +204, +206, +210, +216, +217, +218, +222, +260, +265, and/or +274 according to the numbering of *Bacillus amyloliquefaciens* subtilisin, as described in WO95/10591 and in the patent application of C. Ghosh, et al, "Bleaching

Compositions Comprising Protease Enzymes" having US Serial No. 08/322,677, filed October 13, 1994. Also suitable is a carbonyl hydrolase variant of the protease described in WO95/10591, having an amino acid sequence derived by replacement of a plurality of amino acid residues replaced in the precursor enzyme corresponding to position +210 in combination with one or more of the following residues : +33, +62, +67, +76, +100, +101, +103, +104, +107, +128, +129, +130, +132, +135, +156, +158, +164, +166, +167, +170, +209, +215, +217, +218, and +222, where the numbered position corresponds to naturally-occurring subtilisin from *Bacillus amyloliquefaciens* or to equivalent amino acid residues in other carbonyl hydrolases or subtilisins, such as *Bacillus lentus* subtilisin (co-pending patent application US Serial No. 60/048,550, filed June 04, 1997).

Also preferred proteases are multiply-substituted protease variants. These protease variants comprise a substitution of an amino acid residue with another naturally occurring amino acid residue at an amino acid residue position corresponding to position 103 of *Bacillus amyloliquefaciens* subtilisin in combination with a substitution of an amino acid residue positions corresponding to positions 1, 3, 4, 8, 9, 10, 12, 13, 16, 17, 18, 19, 20, 21, 22, 24, 27, 33, 37, 38, 42, 43, 48, 55, 57, 58, 61, 62, 68, 72, 75, 76, 77, 78, 79, 86, 87, 89, 97, 98, 99, 101, 102, 104, 106, 107, 109, 111, 114, 116, 117, 119, 121, 123, 126, 128, 130, 131, 133, 134, 137, 140, 141, 142, 146, 147, 158, 159, 160, 166, 167, 170, 173, 174, 177, 181, 182, 183, 184, 185, 188, 192, 194, 198, 203, 204, 205, 206, 209, 210, 211, 212, 213, 214, 215, 216, 217, 218, 222, 224, 227, 228, 230, 232, 236, 237, 238, 240, 242, 243, 244, 245, 246, 247, 248, 249, 251, 252, 253, 254, 255, 256, 257, 258, 259, 260, 261, 262, 263, 265, 268, 269, 270, 271, 272, 274 and 275 of *Bacillus amyloliquefaciens* subtilisin; wherein when said protease variant includes a substitution of amino acid residues at positions corresponding to positions 103 and 76, there is also a substitution of an amino acid residue at one or more amino acid residue positions other than amino acid residue positions corresponding to positions 27, 99, 101, 104, 107, 109, 123, 128, 166, 204, 206, 210, 216, 217, 218, 222, 260, 265 or 274 of *Bacillus amyloliquefaciens* subtilisin and/or multiply-substituted protease variants comprising a substitution of an amino acid residue with another naturally occurring amino acid residue at one or more amino acid residue positions corresponding to positions 62, 212, 230, 232, 252 and 257 of *Bacillus amyloliquefaciens* subtilisin as described in PCT application Nos.

PCT/US98/22588, PCT/US98/22482 and PCT/US98/22486 all filed on October 23, 1998 from The Procter & Gamble Company.

Also suitable for the present invention are proteases described in patent applications EP 251 446 and WO 91/06637, protease BLAP® described in WO91/02792 and their variants described in WO 95/23221.

See also a high pH protease from *Bacillus* sp. NCIMB 40338 described in WO 93/18140 A to Novo. Enzymatic detergents comprising protease, one or more other enzymes, and a reversible protease inhibitor are described in WO 92/03529 A to Novo. When desired, a protease having decreased adsorption and increased hydrolysis is available as described in WO 95/07791 to Procter & Gamble. A recombinant trypsin-like protease for detergents suitable herein is described in WO 94/25583 to Novo. Other suitable proteases are described in EP 516 200 by Unilever.

The proteolytic enzymes are incorporated in the detergent compositions of the present invention a level of from 0.0001% to 2%, preferably from 0.001% to 0.2%, more preferably from 0.005% to 0.1% pure enzyme by weight of the composition.

The cellulases without naturally occurring CBDs or with a having a relative binding constant ( $K_r$ -a) for binding to amorphous cellulose lower than 2.4/gcellulose, include both bacterial or fungal cellulases. Preferably, they will have a pH optimum of between 5 and 12 and an activity above 50 CEVU (Cellulose Viscosity Unit). Suitable cellulases are disclosed in U.S. Patent 4,435,307, Barbesgoard et al, J61078384 and WO96/02653 which discloses fungal cellulase produced respectively from *Humicola insolens*, *Trichoderma*, *Thielavia* and *Sporotrichum*. EP 739 982 describes cellulases isolated from novel *Bacillus* species. Suitable cellulases are also disclosed in GB-A-2.075.028; GB-A-2.095.275; DE-OS-2.247.832 and WO95/26398.

Examples of such cellulases are cellulases produced by a strain of *Humicola insolens* (*Humicola grisea* var. *thermoidea*), particularly the *Humicola* strain DSM 1800. Other suitable cellulases are cellulases originated from *Humicola insolens* having a molecular weight of about 50KDa, an isoelectric point of 5.5 and containing 415 amino acids; and a ~43kD endoglucanase derived from *Humicola insolens*, DSM 1800, exhibiting cellulase activity; a preferred endoglucanase component has the amino acid sequence disclosed in PCT Patent Application

No. WO 91/17243. Also suitable cellulases are the EGIII cellulases from *Trichoderma longibrachiatum* described in WO94/21801, Genencor, published September 29, 1994. Especially suitable cellulases are the cellulases having color care benefits. Examples of such cellulases are cellulases described in European patent application No. 91202879.2, filed November 6, 1991 (Novo). Carezyme and Celluzyme (Novo Nordisk A/S) are especially useful. See also WO91/17244 and WO91/21801. Other suitable cellulases for fabric care and/or cleaning properties are described in WO96/34092, WO96/17994 and WO95/24471.

Said cellulases are normally incorporated in the detergent composition at levels from 0.0001% to 2% of pure enzyme by weight of the detergent composition.

Peroxidase enzymes are used in combination with oxygen sources, e.g. percarbonate, perborate, persulfate, hydrogen peroxide, etc and with a phenolic substrate as bleach enhancing molecule. They are used for "solution bleaching", i.e. to prevent transfer of dyes or pigments removed from substrates during wash operations to other substrates in the wash solution. Peroxidase enzymes are known in the art, and include, for example, horseradish peroxidase, ligninase and haloperoxidase such as chloro- and bromo-peroxidase. Peroxidase-containing detergent compositions are disclosed, for example, in PCT International Application WO 89/099813, WO89/09813 and in European Patent application EP No. 91202882.6, filed on November 6, 1991 and EP No. 96870013.8, filed February 20, 1996. Also suitable is the laccase enzyme.

Enhancers are generally comprised at a level of from 0.1% to 5% by weight of total composition. Preferred enhancers are substituted phenothiazine and phenoxazine 10-Phenothiazinepropionic acid (PPT), 10-ethylphenothiazine-4-carboxylic acid (EPC), 10-phenoxazinepropionic acid (POP) and 10-methylphenoxazine (described in WO 94/12621) and substituted syringates (C3-C5 substituted alkyl syringates) and phenols. Sodium percarbonate or perborate are preferred sources of hydrogen peroxide.

Said peroxidases are normally incorporated in the detergent composition at levels from 0.0001% to 2% of pure enzyme by weight of the detergent composition.

Other enzymes that can be included in the detergent compositions of the present invention include lipases. Suitable lipase enzymes for detergent usage

include those produced by microorganisms of the *Pseudomonas* group, such as *Pseudomonas stutzeri* ATCC 19.154, as disclosed in British Patent 1,372,034. Suitable lipases include those which show a positive immunological cross-reaction with the antibody of the lipase, produced by the microorganism *Pseudomonas fluorescent* IAM 1057. This lipase is available from Amano Pharmaceutical Co. Ltd., Nagoya, Japan, under the trade name Lipase P "Amano," hereinafter referred to as "Amano-P". Other suitable commercial lipases include Amano-CES, lipases ex *Chromobacter viscosum*, e.g. *Chromobacter viscosum* var. *lipolyticum* NRRLB 3673 from Toyo Jozo Co., Tagata, Japan; *Chromobacter viscosum* lipases from U.S. Biochemical Corp., U.S.A. and Disoynth Co., The Netherlands, and lipases ex *Pseudomonas gladioli*. Especially suitable lipases are lipases such as M1 Lipase<sup>R</sup> and Lipomax<sup>R</sup> (Gist-Brocades) and Lipolase<sup>R</sup> and Lipolase Ultra<sup>R</sup>(Novo) which have found to be very effective when used in combination with the compositions of the present invention. Also suitable are the lipolytic enzymes described in EP 258 068, WO 92/05249 and WO 95/22615 by Novo Nordisk and in WO 94/03578, WO 95/35381 and WO 96/00292 by Unilever.

Also suitable are cutinases [EC 3.1.1.50] which can be considered as a special kind of lipase, namely lipases which do not require interfacial activation. Addition of cutinases to detergent compositions have been described in e.g. WO-A-88/09367 (Genencor); WO 90/09446 (Plant Genetic System) and WO 94/14963 and WO 94/14964 (Unilever).

The lipases and/or cutinases are normally incorporated in the detergent composition at levels from 0.0001% to 2% of pure enzyme by weight of the detergent composition.

Amylases ( $\alpha$  and/or  $\beta$ ) can be included for removal of carbohydrate-based stains. WO94/02597, Novo Nordisk A/S published February 03, 1994, describes cleaning compositions which incorporate mutant amylases. See also WO95/10603, Novo Nordisk A/S, published April 20, 1995. Other amylases known for use in cleaning compositions include both  $\alpha$ - and  $\beta$ -amylases.  $\alpha$ -Amylases are known in the art and include those disclosed in US Pat. no. 5,003,257; EP 252,666; WO/91/00353; FR 2,676,456; EP 285,123; EP 525,610; EP 368,341; and British Patent specification no. 1,296,839 (Novo). Other suitable amylases are stability-enhanced amylases described in WO94/18314, published August 18, 1994 and WO96/05295, Genencor, published February

22, 1996 and amylase variants having additional modification in the immediate parent available from Novo Nordisk A/S, disclosed in WO 95/10603, published April 95. Also suitable are amylases described in EP 277 216, WO95/26397 and WO96/23873 (all by Novo Nordisk).

Examples of commercial  $\alpha$ -amylases products are Purafect Ox Am<sup>®</sup> from Genencor and Termamyl<sup>®</sup>, Ban<sup>®</sup>, Fungamyl<sup>®</sup> and Duramyl<sup>®</sup>, all available from Novo Nordisk A/S Denmark. WO95/26397 describes other suitable amylases:  $\alpha$ -amylases characterised by having a specific activity at least 25% higher than the specific activity of Termamyl<sup>®</sup> at a temperature range of 25°C to 55°C and at a pH value in the range of 8 to 10, measured by the Phadebas<sup>®</sup>  $\alpha$ -amylase activity assay. Suitable are variants of the above enzymes, described in WO96/23873 (Novo Nordisk). Other amylolytic enzymes with improved properties with respect to the activity level and the combination of thermostability and a higher activity level are described in WO95/35382.

The amylolytic enzymes are incorporated in the detergent compositions of the present invention a level of from 0.0001% to 2%, preferably from 0.00018% to 0.06%, more preferably from 0.00024% to 0.048% pure enzyme by weight of the composition.

The above-mentioned enzymes may be of any suitable origin, such as vegetable, animal, bacterial, fungal and yeast origin. Origin can further be mesophilic or extremophilic (psychrophilic, psychrotrophic, thermophilic, barophilic, alkalophilic, acidophilic, halophilic, etc.). Purified or non-purified forms of these enzymes may be used. Also included by definition, are mutants of native enzymes. Mutants can be obtained e.g. by protein and/or genetic engineering, chemical and/or physical modifications of native enzymes. Common practice as well is the expression of the enzyme via host organisms in which the genetic material responsible for the production of the enzyme has been cloned.

Said enzymes are normally incorporated in the detergent composition at levels from 0.0001% to 2% of pure enzyme by weight of the detergent composition. The enzymes can be added as separate single ingredients (prills, granulates, stabilized liquids, etc. containing one enzyme) or as mixtures of two or more enzymes (e.g. cogramulates).



Other suitable detergent ingredients that can be added are enzyme oxidation scavengers which are described in Copending European Patent application 92870018.6 filed on January 31, 1992. Examples of such enzyme oxidation scavengers are ethoxylated tetraethylene polyamines.

A range of enzyme materials and means for their incorporation into synthetic detergent compositions is also disclosed in WO 9307263 A and WO 9307260 A to Genencor International, WO 8908694 A to Novo, and U.S. 3,553,139, January 5, 1971 to McCarty et al. Enzymes are further disclosed in U.S. 4,101,457, Place et al, July 18, 1978, and in U.S. 4,507,219, Hughes, March 26, 1985. Enzyme materials useful for liquid detergent formulations, and their incorporation into such formulations, are disclosed in U.S. 4,261,868, Hora et al, April 14, 1981. Enzymes for use in detergents can be stabilised by various techniques. Enzyme stabilisation techniques are disclosed and exemplified in U.S. 3,600,319, August 17, 1971, Gedge et al, EP 199,405 and EP 200,586, October 29, 1986, Venegas. Enzyme stabilisation systems are also described, for example, in U.S. 3,519,570. A useful *Bacillus*, sp. AC13 giving proteases, xylanases and cellulases, is described in WO 9401532 A to Novo.

#### ***Colour care and fabric care benefits***

Technologies which provide a type of colour care benefit can also be included. Examples of these technologies are metallo catalysts for colour maintenance. Such metallo catalysts are described in copending European Patent Application No. 92870181.2. Dye fixing agents, polyolefin dispersion for anti-wrinkles and improved water absorbancy, perfume and amino-functional polymer for color care treatment and perfume substantivity are further examples of colour care / fabric care technologies and are described in the co-pending Patent Application No. 96870140.9, filed November 07, 1996.

Fabric softening agents can also be incorporated into laundry detergent and/or fabric care compositions in accordance with the present invention. These agents may be inorganic or organic in type. Inorganic softening agents are exemplified by the smectite clays disclosed in GB-A-1 400 898 and in USP 5,019,292. Organic fabric softening agents include the water insoluble tertiary amines as disclosed in GB-A1 514 276 and EP-B0 011 340 and their combination with

mono C12-C14 quaternary ammonium salts are disclosed in EP-B-0 026 527 and EP-B-0 026 528 and di-long-chain amides as disclosed in EP-B-0 242 919. Other useful organic ingredients of fabric softening systems include high molecular weight polyethylene oxide materials as disclosed in EP-A-0 299 575 and 0 313 146.

Preferred fabric softening agent for the purpose of the present invention is a smectite clay. It has been surprisingly found that the laundry detergent and/or fabric care compositions of the present invention further comprising a smectite clay, achieve improved fabric care, including improved anti-wrinkle, anti-bobbling and anti-shrinkage properties to fabrics, as well as provide enhanced static control, fabric softness, colour appearance and fabric anti-wear properties and benefits while preventing tensile strength loss.

Levels of smectite clay are normally in the range from 2% to 20%, more preferably from 5% to 15% by weight, with the material being added as a dry mixed component to the remainder of the formulation. Organic fabric softening agents such as the water-insoluble tertiary amines or dilong chain amide materials are incorporated at levels of from 0.5% to 5% by weight, normally from 1% to 3% by weight whilst the high molecular weight polyethylene oxide materials and the water soluble cationic materials are added at levels of from 0.1% to 2%, normally from 0.15% to 1.5% by weight. These materials are normally added to the spray dried portion of the composition, although in some instances it may be more convenient to add them as a dry mixed particulate, or spray them as molten liquid on to other solid components of the composition.

### ***Bleaching agent***

Other detergent ingredients that can be included in the laundry detergent and/or fabric care compositions of the present invention include bleaching agents.

Bleaching agents include hydrogen peroxide, PB1, PB4 and percarbonate with a particle size of 400-800 microns. These bleaching agent components can include one or more oxygen bleaching agents and, depending upon the bleaching agent chosen, one or more bleach activators. When present oxygen bleaching compounds will typically be present at levels of from about 1% to about 25%.

The bleaching agent component for use herein can be any of the bleaching agents useful for cleaning compositions including oxygen bleaches as well as others known in the art. The bleaching agent suitable for the present invention can be an activated or non-activated bleaching agent.

One category of oxygen bleaching agent that can be used encompasses percarboxylic acid bleaching agents and salts thereof. Suitable examples of this class of agents include magnesium monoperoxyphthalate hexahydrate, the magnesium salt of meta-chloro perbenzoic acid, 4-nonylamino-4-oxoperoxybutyric acid and diperoxydodecanedioic acid. Such bleaching agents are disclosed in U.S. Patent 4,483,781, U.S. Patent Application 740,446, European Patent Application 0,133,354 and U.S. Patent 4,412,934. Highly preferred bleaching agents also include 6-nonylamino-6-oxoperoxyacaproic acid as described in U.S. Patent 4,634,551.

Another category of bleaching agents that can be used encompasses the halogen bleaching agents. Examples of hypohalite bleaching agents, for example, include trichloro isocyanuric acid and the sodium and potassium dichloroisocyanurates and N-chloro and N-bromo alkane sulphonamides. Such materials are normally added at 0.5-10% by weight of the finished product, preferably 1-5% by weight.

The hydrogen peroxide releasing agents can be used in combination with bleach activators such as tetraacetythylenediamine (TAED), nonanoyloxybenzenesulfonate (NOBS, described in US 4,412,934), 3,5-trimethylhexanoyloxybenzenesulfonate (ISONOBS, described in EP 120,591) or pentaacetylglucose (PAG) or Phenolsulfonate ester of N-nonanoyl-6-aminocaproic acid (NACA-OBS, described in WO94/28106), which are perhydrolyzed to form a peracid as the active bleaching species, leading to improved bleaching effect. Also suitable activators are acylated citrate esters such as disclosed in Copending European Patent Application No. 91870207.7.

Useful bleaching agents, including peroxyacids and bleaching systems comprising bleach activators and peroxygen bleaching compounds for use in detergent compositions according to the invention are described in our co-

pending applications USSN 08/136,626, PCT/US95/07823, WO95/27772, WO95/27773, WO95/27774 and WO95/27775.

The hydrogen peroxide may also be present by adding an enzymatic system (i.e., an enzyme and a substrate therefore) which is capable of generating hydrogen peroxide at the beginning or during the washing and/or rinsing process. Such enzymatic systems are disclosed in EP Patent Application 91202655.6 filed October 9, 1991.

Metal-containing catalysts for use in bleach compositions, include cobalt-containing catalysts such as Pentaamine acetate cobalt(III) salts and manganese-containing catalysts such as those described in EPA 549 271; EPA 549 272; EPA 458 397; US 5,246,621; EPA 458 398; US 5,194,416 and US 5,114,611. Bleaching composition comprising a peroxy compound, a manganese-containing bleach catalyst and a chelating agent is described in the patent application No 94870206.3.

Bleaching agents other than oxygen bleaching agents are also known in the art and can be utilized herein. One type of non-oxygen bleaching agent of particular interest includes photoactivated bleaching agents such as the sulfonated zinc and/or aluminum phthalocyanines. These materials can be deposited upon the substrate during the washing process. Upon irradiation with light, in the presence of oxygen, such as by hanging clothes out to dry in the daylight, the sulfonated zinc phthalocyanine is activated and, consequently, the substrate is bleached. Preferred zinc phthalocyanine and a photoactivated bleaching process are described in U.S. Patent 4,033,718. Typically, detergent compositions will contain about 0.025% to about 1.25%, by weight, of sulfonated zinc phthalocyanine.

### ***Builder system***

The compositions according to the present invention may further comprise a builder system. It has been surprisingly found that said laundry detergent and/or fabric care compositions further comprising a builder, achieve improved fabric care, including improved anti-wrinkle, anti-bobbling and anti-shrinkage properties to fabrics, as well as provide enhanced static control, fabric softness, colour

appearance and fabric anti-wear properties and benefits and improved fabric cleaning while preventing tensile strength loss. Preferred builders are zeolite A and sodium tripolyphosphate.

Any conventional builder system is suitable for use herein including aluminosilicate materials, silicates, polycarboxylates, alkyl- or alkenyl-succinic acid and fatty acids, materials such as ethylenediamine tetraacetate, diethylene triamine pentamethyleneacetate, metal ion sequestrants such as aminopolyphosphonates, particularly ethylenediamine tetramethylene phosphonic acid and diethylene triamine pentamethylenephosphonic acid. Phosphate builders can also be used herein.

Suitable builders can be an inorganic ion exchange material, commonly an inorganic hydrated aluminosilicate material, more particularly a hydrated synthetic zeolite such as hydrated zeolite A, X, B, HS or MAP.

Another suitable inorganic builder material is layered silicate, e.g. SKS-6 (Hoechst). SKS-6 is a crystalline layered silicate consisting of sodium silicate ( $\text{Na}_2\text{Si}_2\text{O}_5$ ).

Suitable polycarboxylates containing one carboxy group include lactic acid, glycolic acid and ether derivatives thereof as disclosed in Belgian Patent Nos. 831,368, 821,369 and 821,370. Polycarboxylates containing two carboxy groups include the water-soluble salts of succinic acid, malonic acid, (ethylenedioxy) diacetic acid, maleic acid, diglycollic acid, tartaric acid, tartronic acid and fumaric acid, as well as the ether carboxylates described in German Offenlegenschrift 2,446,686, and 2,446,687 and U.S. Patent No. 3,935,257 and the sulfinyl carboxylates described in Belgian Patent No. 840,623. Polycarboxylates containing three carboxy groups include, in particular, water-soluble citrates, aconitrates and citraconates as well as succinate derivatives such as the carboxymethyloxysuccinates described in British Patent No. 1,379,241, lactoxysuccinates described in Netherlands Application 7205873, and the oxypolycarboxylate materials such as 2-oxa-1,1,3-propane tricarboxylates described in British Patent No. 1,387,447.

Polycarboxylates containing four carboxy groups include oxydisuccinates disclosed in British Patent No. 1,261,829, 1,1,2,2-ethane tetracarboxylates, 1,1,3,3-propane tetracarboxylates and 1,1,2,3-propane tetracarboxylates.

Polycarboxylates containing sulfo substituents include the sulfosuccinate derivatives disclosed in British Patent Nos. 1,398,421 and 1,398,422 and in U.S. Patent No. 3,936,448, and the sulfonated pyrolysed citrates described in British Patent No. 1,082,179, while polycarboxylates containing phosphone substituents are disclosed in British Patent No. 1,439,000.

Alicyclic and heterocyclic polycarboxylates include cyclopentane-cis,cis,cis-tetracarboxylates, cyclopentadienide pentacarboxylates, 2,3,4,5-tetrahydro-furan - cis, cis, cis-tetracarboxylates, 2,5-tetrahydro-furan -cis - dicarboxylates, 2,2,5,5-tetrahydrofuran - tetracarboxylates, 1,2,3,4,5,6-hexane -hexacarboxylates and and carboxymethyl derivatives of polyhydric alcohols such as sorbitol, mannitol and xylitol. Aromatic poly-carboxylates include mellitic acid, pyromellitic acid and the phthalic acid derivatives disclosed in British Patent No. 1,425,343.

Of the above, the preferred polycarboxylates are hydroxycarboxylates containing up to three carboxy groups per molecule, more particularly citrates.

Preferred builder systems for use in the present compositions include a mixture of a water-insoluble aluminosilicate builder such as zeolite A or of a layered silicate (SKS-6), and a water-soluble carboxylate chelating agent such as citric acid.

Preferred builder systems include a mixture of a water-insoluble aluminosilicate builder such as zeolite A, and a watersoluble carboxylate chelating agent such as citric acid. Preferred builder systems for use in liquid detergent compositions of the present invention are soaps and polycarboxylates.

Other builder materials that can form part of the builder system for use in granular compositions include inorganic materials such as alkali metal carbonates, bicarbonates, silicates, and organic materials such as the organic phosphonates, amino polyalkylene phosphonates and amino polycarboxylates.

Other suitable water-soluble organic salts are the homo- or co-polymeric acids or their salts, in which the polycarboxylic acid comprises at least two carboxyl radicals separated from each other by not more than two carbon atoms.

Polymers of this type are disclosed in GB-A-1,596,756. Examples of such salts are polyacrylates of MW 2000-5000 and their copolymers with maleic anhydride, such copolymers having a molecular weight of from 20,000 to 70,000, especially about 40,000.

Detergency builder salts are normally included in amounts of from 5% to 80% by weight of the composition preferably from 10% to 70% and most usually from 30% to 60% by weight.

### ***Chelating Agents***

The laundry detergent and/or fabric care compositions herein may also optionally contain one or more iron and/or manganese chelating agents. Such chelating agents can be selected from the group consisting of amino carboxylates, amino phosphonates, polyfunctionally-substituted aromatic chelating agents and mixtures therein, all as hereinafter defined. Without intending to be bound by theory, it is believed that the benefit of these materials is due in part to their exceptional ability to remove iron and manganese ions from washing solutions by formation of soluble chelates.

Amino carboxylates useful as optional chelating agents include ethylenediaminetetracetates, N-hydroxyethylethylenediaminetriacetates, nitrilotriacetates, ethylenediamine tetrapropionates, triethylenetetraaminehexacetates, diethylenetriaminepentaacetates, and ethanoldiglycines, alkali metal, ammonium, and substituted ammonium salts therein and mixtures therein.

Amino phosphonates are also suitable for use as chelating agents in the compositions of the invention when at least low levels of total phosphorus are permitted in detergent compositions, and include ethylenediaminetetrakis (methylenephosphonates) as DEQUEST. Preferred, these amino phosphonates do not contain alkyl or alkenyl groups with more than about 6 carbon atoms.

Polyfunctionally-substituted aromatic chelating agents are also useful in the compositions herein. See U.S. Patent 3,812,044, issued May 21, 1974, to Connor et al. Preferred compounds of this type in acid form are dihydroxydisulfobenzenes such as 1,2-dihydroxy-3,5-disulfobenzene.

A preferred biodegradable chelator for use herein is ethylenediamine disuccinate ("EDDS"), especially the [S,S] isomer as described in U.S. Patent 4,704,233, November 3, 1987, to Hartman and Perkins.

The compositions herein may also contain water-soluble methyl glycine diacetic acid (MGDA) salts (or acid form) as a chelant or co-builder useful with, for example, insoluble builders such as zeolites, layered silicates and the like.

If utilized, these chelating agents will generally comprise from about 0.1% to about 15% by weight of the detergent compositions herein. More preferably, if utilized, the chelating agents will comprise from about 0.1% to about 3.0% by weight of such compositions.

### ***Suds suppressor***

Another optional ingredient is a suds suppressor, exemplified by silicones, and silica-silicone mixtures. Silicones can be generally represented by alkylated polysiloxane materials while silica is normally used in finely divided forms exemplified by silica aerogels and xerogels and hydrophobic silicas of various types. These materials can be incorporated as particulates in which the suds suppressor is advantageously releasably incorporated in a water-soluble or water-dispersible, substantially non-surface-active detergent impermeable carrier. Alternatively the suds suppressor can be dissolved or dispersed in a liquid carrier and applied by spraying on to one or more of the other components.

A preferred silicone suds controlling agent is disclosed in Bartollota et al. U.S. Patent 3 933 672. Other particularly useful suds suppressors are the self-emulsifying silicone suds suppressors, described in German Patent Application DTOS 2 646 126 published April 28, 1977. An example of such a compound is DC-544, commercially available from Dow Corning, which is a siloxane-glycol copolymer. Especially preferred suds controlling agent are the suds suppressor system comprising a mixture of silicone oils and 2-alkyl-alkanols. Suitable 2-alkyl-alkanols are 2-butyl-octanol which are commercially available under the trade name Isofol 12 R.



Such suds suppressor system are described in Copending European Patent application N 92870174.7 filed 10 November, 1992.

Especially preferred silicone suds controlling agents are described in Copending European Patent application N°92201649.8. Said compositions can comprise a silicone/silica mixture in combination with fumed nonporous silica such as Aerosil<sup>R</sup>.

The suds suppressors described above are normally employed at levels of from 0.001% to 2% by weight of the composition, preferably from 0.01% to 1% by weight.

### ***Others***

Other components such as soil-suspending agents, soil-release agents, optical brighteners, abrasives, bactericides, tarnish inhibitors, coloring agents, and/or encapsulated or non-encapsulated perfumes may be employed.

Especially suitable encapsulating materials are water soluble capsules which consist of a matrix of polysaccharide and polyhydroxy compounds such as described in GB 1,464,616.

Other suitable water soluble encapsulating materials comprise dextrans derived from ungelatinized starch acid-esters of substituted dicarboxylic acids such as described in US 3,455,838. These acid-ester dextrans are, preferably, prepared from such starches as waxy maize, waxy sorghum, sago, tapioca and potato. Suitable examples of said encapsulating materials include N-Lok manufactured by National Starch. The N-Lok encapsulating material consists of a modified maize starch and glucose. The starch is modified by adding monofunctional substituted groups such as octenyl succinic acid anhydride.

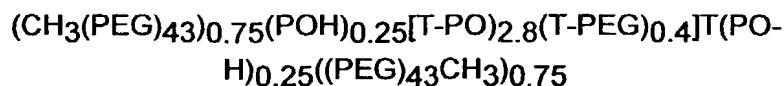
Antiredeposition and soil suspension agents suitable herein include cellulose derivatives such as methylcellulose, carboxymethylcellulose and hydroxyethylcellulose, and homo- or co-polymeric polycarboxylic acids or their salts. Polymers of this type include the polyacrylates and maleic anhydride-acrylic acid copolymers previously mentioned as builders, as well as copolymers of maleic anhydride with ethylene, methylvinyl ether or methacrylic acid, the

maleic anhydride constituting at least 20 mole percent of the copolymer. These materials are normally used at levels of from 0.5% to 10% by weight, more preferably from 0.75% to 8%, most preferably from 1% to 6% by weight of the composition.

Preferred optical brighteners are anionic in character, examples of which are disodium 4,4'-bis-(2-diethanolamino-4-anilino -s- triazin-6-ylamino)stilbene-2:2' disulphonate, disodium 4, - 4'-bis-(2-morpholino-4-anilino-s-triazin-6-ylamino)stilbene-2:2' - disulphonate, disodium 4,4' - bis-(2,4-dianilino-s-triazin-6-ylamino)stilbene-2:2' - disulphonate, monosodium 4,4'' -bis-(2,4-dianilino-s-triazin-6 ylamino)stilbene-2-sulphonate, disodium 4,4' -bis-(2-anilino-4-(N-methyl-N-2-hydroxyethylamino)-s-triazin-6-ylamino)stilbene-2,2' - disulphonate, disodium 4,4' -bis-(4-phenyl-2,1,3-triazol-2-yl)-stilbene-2,2' disulphonate, disodium 4,4'bis(2-anilino-4-(1-methyl-2-hydroxyethylamino)-s-triazin-6- ylamino)stilbene-2,2'disulphonate, sodium 2(stilbyl-4''-(naphtho-1',2':4,5)-1,2,3 - triazole-2''-sulphonate and 4,4'-bis(2-sulphostyryl)biphenyl. Highly preferred brighteners are the specific brighteners of copending European Patent application No. 95201943.8.

Other useful polymeric materials are the polyethylene glycols, particularly those of molecular weight 1000-10000, more particularly 2000 to 8000 and most preferably about 4000. These are used at levels of from 0.20% to 5% more preferably from 0.25% to 2.5% by weight. These polymers and the previously mentioned homo- or co-polymeric polycarboxylate salts are valuable for improving whiteness maintenance, fabric ash deposition, and cleaning performance on clay, proteinaceous and oxidizable soils in the presence of transition metal impurities.

Soil release agents useful in compositions of the present invention are conventionally copolymers or terpolymers of terephthalic acid with ethylene glycol and/or propylene glycol units in various arrangements. Examples of such polymers are disclosed in the commonly assigned US Patent Nos. 4116885 and 4711730 and European Published Patent Application No. 0 272 033. A particular preferred polymer in accordance with EP-A-0 272 033 has the formula



where PEG is  $-(\text{OC}_2\text{H}_4)_n\text{O}-$ , PO is  $(\text{OC}_3\text{H}_6\text{O})$  and T is  $(\text{pOC}_6\text{H}_4\text{CO})$ .

Also very useful are modified polyesters as random copolymers of dimethyl terephthalate, dimethyl sulfoisophthalate, ethylene glycol and 1-2 propane diol, the end groups consisting primarily of sulphobenzoate and secondarily of mono esters of ethylene glycol and/or propane-diol. The target is to obtain a polymer capped at both end by sulphobenzoate groups, "primarily", in the present context most of said copolymers herein will be end-capped by sulphobenzoate groups. However, some copolymers will be less than fully capped, and therefore their end groups may consist of monoester of ethylene glycol and/or propane 1-2 diol, thereof consist "secondarily" of such species.

The selected polyesters herein contain about 46% by weight of dimethyl terephthalic acid, about 16% by weight of propane -1.2 diol, about 10% by weight ethylene glycol about 13% by weight of dimethyl sulfobenzoic acid and about 15% by weight of sulfoisophthalic acid, and have a molecular weight of about 3.000. The polyesters and their method of preparation are described in detail in EPA 311 342.

It is well known in the art that free chlorine in tap water rapidly deactivates the enzymes comprised in detergent compositions. Therefore, using chlorine scavenger such as perborate, ammonium sulfate, sodium sulphite or polyethyleneimine at a level above 0.1% by weight of total composition, in the formulas will provide improved through the wash stability of the detergent enzymes. Compositions comprising chlorine scavenger are described in the European patent application 92870018.6 filed January 31, 1992.

Alkoxyated polycarboxylates such as those prepared from polyacrylates are useful herein to provide additional grease removal performance. Such materials are described in WO 91/08281 and PCT 90/01815 at p. 4 et seq., incorporated herein by reference. Chemically, these materials comprise polyacrylates having one ethoxy side-chain per every 7-8 acrylate units. The side-chains are of the formula  $-(\text{CH}_2\text{CH}_2\text{O})_m(\text{CH}_2)_n\text{CH}_3$  wherein m is 2-3 and n is 6-12. The side-

chains are ester-linked to the polyacrylate "backbone" to provide a "comb" polymer type structure. The molecular weight can vary, but is typically in the range of about 2000 to about 50,000. Such alkoxylated polycarboxylates can comprise from about 0.05% to about 10%, by weight, of the compositions herein.

### ***Dispersants***

The laundry detergent and/or fabric care composition of the present invention can also contain dispersants: Suitable water-soluble organic salts are the homo- or co-polymeric acids or their salts, in which the polycarboxylic acid comprises at least two carboxyl radicals separated from each other by not more than two carbon atoms.

Polymers of this type are disclosed in GB-A-1,596,756. Examples of such salts are polyacrylates of MW 2000-5000 and their copolymers with maleic anhydride, such copolymers having a molecular weight of from 1,000 to 100,000.

Especially, copolymer of acrylate and methylacrylate such as the 480N having a molecular weight of 4000, at a level from 0.5-20% by weight of composition can be added in the laundry detergent and/or fabric care compositions of the present invention.

The compositions of the invention may contain a lime soap peptiser compound, which has preferably a lime soap dispersing power (LSDP), as defined hereinafter of no more than 8, preferably no more than 7, most preferably no more than 6. The lime soap peptiser compound is preferably present at a level from 0% to 20% by weight.

A numerical measure of the effectiveness of a lime soap peptiser is given by the lime soap dispersant power (LSDP) which is determined using the lime soap dispersant test as described in an article by H.C. Borghetty and C.A. Bergman, J. Am. Oil. Chem. Soc., volume 27, pages 88-90, (1950). This lime soap dispersion test method is widely used by practitioners in this art field being referred to, for example, in the following review articles; W.N. Linfield, Surfactant science Series, Volume 7, page 3; W.N. Linfield, Tenside surf. det., volume 27,

pages 159-163, (1990); and M.K. Nagarajan, W.F. Masler, *Cosmetics and Toiletries*, volume 104, pages 71-73, (1989). The LSDP is the % weight ratio of dispersing agent to sodium oleate required to disperse the lime soap deposits formed by 0.025g of sodium oleate in 30ml of water of 333ppm  $\text{CaCO}_3$  (Ca:Mg=3:2) equivalent hardness.

Surfactants having good lime soap peptiser capability will include certain amine oxides, betaines, sulfobetaines, alkyl ethoxysulfates and ethoxylated alcohols.

Exemplary surfactants having a LSDP of no more than 8 for use in accord with the present invention include  $\text{C}_{16}$ - $\text{C}_{18}$  dimethyl amine oxide,  $\text{C}_{12}$ - $\text{C}_{18}$  alkyl ethoxysulfates with an average degree of ethoxylation of from 1-5, particularly  $\text{C}_{12}$ - $\text{C}_{15}$  alkyl ethoxysulfate surfactant with a degree of ethoxylation of amount 3 (LSDP=4), and the  $\text{C}_{14}$ - $\text{C}_{15}$  ethoxylated alcohols with an average degree of ethoxylation of either 12 (LSDP=6) or 30, sold under the tradenames Lutensol A012 and Lutensol A030 respectively, by BASF GmbH.

Polymeric lime soap peptisers suitable for use herein are described in the article by M.K. Nagarajan, W.F. Masler, to be found in *Cosmetics and Toiletries*, volume 104, pages 71-73, (1989).

Hydrophobic bleaches such as 4-[N-octanoyl-6-aminohexanoyl]benzene sulfonate, 4-[N-nonanoyl-6-aminohexanoyl]benzene sulfonate, 4-[N-decanoyl-6-aminohexanoyl]benzene sulfonate and mixtures thereof; and nonanoyloxy benzene sulfonate together with hydrophilic / hydrophobic bleach formulations can also be used as lime soap peptisers compounds.

### ***Dye transfer inhibition***

The laundry detergent and/or fabric care compositions of the present invention can preferably include compounds for inhibiting dye transfer from one fabric to another of solubilized and suspended dyes encountered during fabric laundering operations involving colored fabrics. It has been surprisingly found that the laundry detergent and/or fabric care compositions of the present invention further comprising a dye transfer inhibition polymer, achieve improved fabric care, including improved anti-wrinkle, anti-bobbling and anti-shrinkage properties

to fabrics, as well as provide enhanced static control, fabric softness, colour appearance and fabric anti-wear properties and benefits and improved fabric cleaning while preventing tensile strength loss.

### ***Polymeric dye transfer inhibiting agents***

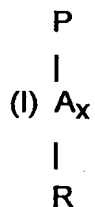
The laundry detergent and/or fabric care compositions according to the present invention may also comprise from 0.001% to 10 %, preferably from 0.01% to 2%, more preferably from 0.05% to 1% by weight of polymeric dye transfer inhibiting agents. Said polymeric dye transfer inhibiting agents are normally incorporated into cleaning compositions in order to inhibit the transfer of dyes from colored fabrics onto fabrics washed therewith. These polymers have the ability to complex or adsorb the fugitive dyes washed out of dyed fabrics before the dyes have the opportunity to become attached to other articles in the wash.

Especially suitable polymeric dye transfer inhibiting agents are polyamine N-oxide polymers, copolymers of N-vinylpyrrolidone and N-vinylimidazole, polyvinylpyrrolidone polymers, polyvinylloxazolidones and polyvinylimidazoles or mixtures thereof.

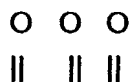
Addition of such polymers also enhances the performance of the enzymes according the invention.

#### **a) Polyamine N-oxide polymers**

The polyamine N-oxide polymers suitable for use contain units having the following structure formula :



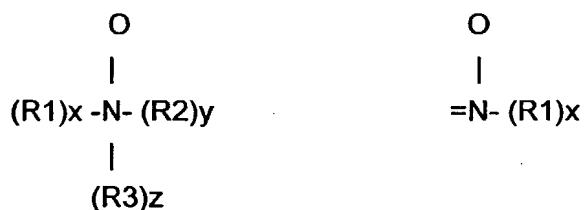
wherein P is a polymerisable unit, whereto the R-N-O group can be attached to or wherein the R-N-O group forms part of the polymerisable unit or a combination of both.



A is NC, CO, C, -O-, -S-, -N- ; x is 0 or 1;

R are aliphatic, ethoxylated aliphatics, aromatic, heterocyclic or alicyclic groups or any combination thereof whereto the nitrogen of the N-O group can be attached or wherein the nitrogen of the N-O group is part of these groups.

The N-O group can be represented by the following general structures :



wherein R1, R2, and R3 are aliphatic groups, aromatic, heterocyclic or alicyclic groups or combinations thereof, x or/and y or/and z is 0 or 1 and wherein the nitrogen of the N-O group can be attached or wherein the nitrogen of the N-O group forms part of these groups.

The N-O group can be part of the polymerisable unit (P) or can be attached to the polymeric backbone or a combination of both.

Suitable polyamine N-oxides wherein the N-O group forms part of the polymerisable unit comprise polyamine N-oxides wherein R is selected from aliphatic, aromatic, alicyclic or heterocyclic groups.

One class of said polyamine N-oxides comprises the group of polyamine N-oxides wherein the nitrogen of the N-O group forms part of the R-group. Preferred polyamine N-oxides are those wherein R is a heterocyclic group such as pyrridine, pyrrole, imidazole, pyrrolidine, piperidine, quinoline, acridine and derivatives thereof.

Another class of said polyamine N-oxides comprises the group of polyamine N-oxides wherein the nitrogen of the N-O group is attached to the R-group.

Other suitable polyamine N-oxides are the polyamine oxides whereto the N-O group is attached to the polymerisable unit.

Preferred class of these polyamine N-oxides are the polyamine N-oxides having the general formula (I) wherein R is an aromatic, heterocyclic or alicyclic groups wherein the nitrogen of the N-O functional group is part of said R group.

Examples of these classes are polyamine oxides wherein R is a heterocyclic compound such as pyridine, pyrrole, imidazole and derivatives thereof.

Another preferred class of polyamine N-oxides are the polyamine oxides having the general formula (I) wherein R are aromatic, heterocyclic or alicyclic groups wherein the nitrogen of the N-O functional group is attached to said R groups.

Examples of these classes are polyamine oxides wherein R groups can be aromatic such as phenyl.

Any polymer backbone can be used as long as the amine oxide polymer formed is water-soluble and has dye transfer inhibiting properties. Examples of suitable polymeric backbones are polyvinyls, polyalkylenes, polyesters, polyethers, polyamide, polyimides, polyacrylates and mixtures thereof.

The amine N-oxide polymers of the present invention typically have a ratio of amine to the amine N-oxide of 10:1 to 1:1000000. However the amount of amine oxide groups present in the polyamine oxide polymer can be varied by appropriate copolymerization or by appropriate degree of N-oxidation. Preferably, the ratio of amine to amine N-oxide is from 2:3 to 1:1000000. More preferably from 1:4 to 1:1000000, most preferably from 1:7 to 1:1000000. The polymers of the present invention actually encompass random or block copolymers where one monomer type is an amine N-oxide and the other monomer type is either an amine N-oxide or not. The amine oxide unit of the polyamine N-oxides has a  $PK_a < 10$ , preferably  $PK_a < 7$ , more preferred  $PK_a < 6$ .

The polyamine oxides can be obtained in almost any degree of polymerisation. The degree of polymerisation is not critical provided the material has the desired water-solubility and dye-suspending power.

Typically, the average molecular weight is within the range of 500 to 1000,000; preferably from 1,000 to 50,000, more preferably from 2,000 to 30,000, most preferably from 3,000 to 20,000.

b) Copolymers of N-vinylpyrrolidone and N-vinylimidazole



The N-vinylimidazole N-vinylpyrrolidone polymers used in the present invention have an average molecular weight range from 5,000-1,000,000, preferably from 5,000-200,000.

Highly preferred polymers for use in detergent compositions according to the present invention comprise a polymer selected from N-vinylimidazole N-vinylpyrrolidone copolymers wherein said polymer has an average molecular weight range from 5,000 to 50,000 more preferably from 8,000 to 30,000, most preferably from 10,000 to 20,000.

The average molecular weight range was determined by light scattering as described in Barth H.G. and Mays J.W. Chemical Analysis Vol 113, "Modern Methods of Polymer Characterization".

Highly preferred N-vinylimidazole N-vinylpyrrolidone copolymers have an average molecular weight range from 5,000 to 50,000; more preferably from 8,000 to 30,000; most preferably from 10,000 to 20,000.

The N-vinylimidazole N-vinylpyrrolidone copolymers characterized by having said average molecular weight range provide excellent dye transfer inhibiting properties while not adversely affecting the cleaning performance of detergent compositions formulated therewith.

The N-vinylimidazole N-vinylpyrrolidone copolymer of the present invention has a molar ratio of N-vinylimidazole to N-vinylpyrrolidone from 1 to 0.2, more preferably from 0.8 to 0.3, most preferably from 0.6 to 0.4 .

#### c) Polyvinylpyrrolidone

The detergent compositions of the present invention may also utilize polyvinylpyrrolidone ("PVP") having an average molecular weight of from about 2,500 to about 400,000, preferably from about 5,000 to about 200,000, more preferably from about 5,000 to about 50,000, and most preferably from about 5,000 to about 15,000. Suitable polyvinylpyrrolidones are commercially available from ISP Corporation, New York, NY and Montreal, Canada under the product names PVP K-15 (viscosity molecular weight of 10,000), PVP K-30 (average molecular weight of 40,000), PVP K-60 (average molecular weight of 160,000), and PVP K-90 (average molecular weight of 360,000). Other suitable polyvinylpyrrolidones which are commercially available from BASF Cooperation include Sokalan HP 165 and Sokalan HP 12; polyvinylpyrrolidones known to

persons skilled in the detergent field (see for example EP-A-262,897 and EP-A-256,696).

d) Polyvinylloxazolidone :

The detergent compositions of the present invention may also utilize polyvinylloxazolidone as a polymeric dye transfer inhibiting agent. Said polyvinylloxazolidones have an average molecular weight of from about 2,500 to about 400,000, preferably from about 5,000 to about 200,000, more preferably from about 5,000 to about 50,000, and most preferably from about 5,000 to about 15,000.

e) Polyvinylimidazole :

The detergent compositions of the present invention may also utilize polyvinylimidazole as polymeric dye transfer inhibiting agent. Said polyvinylimidazoles have an average of about 2,500 to about 400,000, preferably from about 5,000 to about 200,000, more preferably from about 5,000 to about 50,000, and most preferably from about 5,000 to about 15,000.

f) Cross-linked polymers :

Cross-linked polymers are polymers whose backbone are interconnected to a certain degree; these links can be of chemical or physical nature, possibly with active groups in the backbone or on branches; cross-linked polymers have been described in the Journal of Polymer Science, volume 22, pages 1035-1039.

In one embodiment, the cross-linked polymers are made in such a way that they form a three-dimensional rigid structure, which can entrap dyes in the pores formed by the three-dimensional structure. In another embodiment, the cross-linked polymers entrap the dyes by swelling.

Such cross-linked polymers are described in the co-pending patent application 94870213.9

### **Method of washing**

The compositions of the invention may be used in essentially any washing, cleaning and/or fabric care methods, including soaking methods, pre-treatment methods, methods with rinsing steps for which a separate rinse aid composition may be added and post-treatment methods.

The process described herein comprises contacting fabrics with a laundering solution in the usual manner and exemplified hereunder. A conventional laundry method comprises treating soiled fabric with an aqueous liquid having dissolved or dispensed therein an effective amount of the laundry detergent and/or fabric care composition. The process of the invention is conveniently carried out in the course of the cleaning process. The method of cleaning is preferably carried out at 5°C to 95°C, especially between 10°C and 60°C. The pH of the treatment solution is preferably from 7 to 12.

The following examples are meant to exemplify compositions of the present invention, but are not necessarily meant to limit or otherwise define the scope of the invention.

In the detergent compositions, the enzymes levels are expressed by pure enzyme by weight of the total composition and unless otherwise specified, the detergent ingredients are expressed by weight of the total compositions. The abbreviated component identifications therein have the following meanings:

LAS	: Sodium linear C <sub>11-13</sub> alkyl benzene sulphonate.
TAS	: Sodium tallow alkyl sulphate.
C <sub>xy</sub> AS	: Sodium C <sub>1x</sub> - C <sub>1y</sub> alkyl sulfate.
C <sub>xy</sub> SAS	: Sodium C <sub>1x</sub> - C <sub>1y</sub> secondary (2,3) alkyl sulfate.
C <sub>xy</sub> Ez	: C <sub>1x</sub> - C <sub>1y</sub> predominantly linear primary alcohol condensed with an average of z moles of ethylene oxide.
C <sub>xy</sub> EzS	: C <sub>1x</sub> - C <sub>1y</sub> sodium alkyl sulfate condensed with an average of z moles of ethylene oxide.
QAS	: R <sub>2</sub> .N+(CH <sub>3</sub> ) <sub>2</sub> (C <sub>2</sub> H <sub>4</sub> OH) with R <sub>2</sub> = C <sub>12</sub> -C <sub>14</sub> .
QAS 1	: R <sub>2</sub> .N+(CH <sub>3</sub> ) <sub>2</sub> (C <sub>2</sub> H <sub>4</sub> OH) with R <sub>2</sub> = C <sub>8</sub> -C <sub>11</sub> .
APA	: C <sub>8-10</sub> amido propyl dimethyl amine.

Soap	: Sodium linear alkyl carboxylate derived from a 80/20 mixture of tallow and coconut fatty acids.
STS	: Sodium toluene sulphonate.
CFAA	: C <sub>12</sub> -C <sub>14</sub> alkyl N-methyl glucamide.
TFAA	: C <sub>16</sub> -C <sub>18</sub> alkyl N-methyl glucamide.
TPKFA	: C <sub>12</sub> -C <sub>14</sub> topped whole cut fatty acids.
DEQA	: Di-(tallow-oxy-ethyl) dimethyl ammonium chloride.
DEQA (2)	: Di-(soft-tallowyloxyethyl) hydroxyethyl methyl ammonium methylsulfate.
DTDMAMS	: Ditallow dimethyl ammonium methylsulfate.
SDASA	: 1:2 ratio of stearyldimethyl amine:triple-pressed stearic acid.
Silicate	: Amorphous Sodium Silicate (SiO <sub>2</sub> :Na <sub>2</sub> O ratio = 1.6-3.2).
Zeolite A	: Hydrated Sodium Aluminosilicate of formula Na <sub>12</sub> (AlO <sub>2</sub> SiO <sub>2</sub> ) <sub>12</sub> . 27H <sub>2</sub> O having a primary particle size in the range from 0.1 to 10 micrometers (Weight expressed on an anhydrous basis).
Na-SKS-6	: Crystalline layered silicate of formula $\delta$ -Na <sub>2</sub> Si <sub>2</sub> O <sub>5</sub> .
Citrate	: Tri-sodium citrate dihydrate of activity 86.4% with a particle size distribution between 425 and 850 micrometres.
Citric	: Anhydrous citric acid.
Borate	: Sodium borate
Carbonate	: Anhydrous sodium carbonate with a particle size between 200 and 900 micrometres.
Bicarbonate	: Anhydrous sodium hydrogen carbonate with a particle size distribution between 400 and 1200 micrometres.
Sulphate	: Anhydrous sodium sulphate.
Mg Sulphate	: Anhydrous magnesium sulfate.
STPP	: Sodium tripolyphosphate.
TSPP	: Tetrasodium pyrophosphate.
MA/AA	: Random copolymer of 4:1 acrylate/maleate, average molecular weight about 70,000-80,000.
MA/AA 1	: Random copolymer of 6:4 acrylate/maleate, average molecular weight about 10,000.

AA	: Sodium polyacrylate polymer of average molecular weight 4,500.
PB1	: Anhydrous sodium perborate monohydrate of nominal formula $\text{NaBO}_2 \cdot \text{H}_2\text{O}_2$ .
PB4	: Sodium perborate tetrahydrate of nominal formula $\text{NaBO}_2 \cdot 3\text{H}_2\text{O} \cdot \text{H}_2\text{O}_2$ .
Percarbonate	: Anhydrous sodium percarbonate of nominal formula $2\text{Na}_2\text{CO}_3 \cdot 3\text{H}_2\text{O}_2$ .
TAED	: Tetraacetylenediamine.
NOBS	: Nonanoyloxybenzene sulfonate in the form of the sodium salt.
NACA-OBS	: (6-nonamidocaproyl) oxybenzene sulfonate.
DTPA	: Diethylene triamine pentaacetic acid.
HEDP	: 1,1-hydroxyethane diphosphonic acid.
DETPMP	: Diethyltriamine penta (methylene) phosphonate, marketed by Monsanto under the Trade name Dequest 2060.
EDDS	: Ethylenediamine-N,N'-disuccinic acid, (S,S) isomer in the form of its sodium salt
Photoactivated Bleach	: Sulfonated zinc phthalocyanine encapsulated in dextrin soluble polymer.
Photoactivated Bleach 1	: Sulfonated alumino phthalocyanine encapsulated in dextrin soluble polymer.
Protease	: Proteolytic enzyme sold under the tradename Savinase, Alcalase, Durazym by Novo Nordisk A/S, Maxacal, Maxapem sold by Gist-Brocades and proteases described in patents WO91/06637 and/or WO95/10591 and/or EP 251 446.
Amylase	: Amylolytic enzyme sold under the tradename Purafact Ox Am <sup>R</sup> described in WO 94/18314, WO96/05295 sold by Genencor, Termamyl <sup>®</sup> , Fungamyl <sup>®</sup> and Duramyl <sup>®</sup> , all available from Novo Nordisk A/S and those described in WO95/26397.
Lipase	: Lipolytic enzyme sold under the tradename Lipolase, Lipolase Ultra by Novo Nordisk A/S and Lipomax by Gist-Brocades.

- CBD-Cellulase : Cellulytic enzyme core derived from the enzyme sold under the tradename Carezyme by Novo Nordisk A/S linked to the CBD of the CenC cellulase of *Cellulomonas fimi*, either the tandem CBDN1N2 or the single CBDN1 or CBDN2 and/or linked to the CBD from the cellulase E3 of *Thermomonospora fusca*; and/or
- : Cellulytic enzyme core derived from the enzyme sold under the tradename Carezyme by Novo Nordisk A/S linked by the *Humicola Insolens* family 45 cellulase linker to the CBD of the CenC cellulase of *Cellulomonas fimi*, either the tandem CBDN1N2 or the single CBDN1 or CBDN2; and/or
- : Cellulytic enzyme core derived from the enzyme sold under the tradename Carezyme by Novo Nordisk A/S linked by the E3 cellulase *Thermomonospora fusca* linker to the CBD from the cellulase E3 of *Thermomonospora fusca*.
- Cellulase : Cellulytic enzyme sold under the tradename Carezyme, Endolase by Novo Nordisk A/S; and/or Clazinase, Puradex HAI, EGIII cellulase from *Trichoderma reseei* by Genencor.
- CMC : Sodium carboxymethyl cellulose.
- PVP : Polyvinyl polymer, with an average molecular weight of 60,000.
- PVNO : Polyvinylpyridine-N-Oxide, with an average molecular weight of 50,000.
- PVPVI : Copolymer of vinylimidazole and vinylpyrrolidone, with an average molecular weight of 20,000.
- Brightener 1 : Disodium 4,4'-bis(2-sulphostyryl)biphenyl.
- Brightener 2 : Disodium 4,4'-bis(4-anilino-6-morpholino-1.3.5-triazin-2-yl) stilbene-2:2'-disulfonate.
- Silicone antifoam : Polydimethylsiloxane foam controller with siloxane-oxyalkylene copolymer as dispersing agent with a ratio of said foam controller to said dispersing agent of 10:1 to 100:1.

- Suds Suppressor : 12% Silicone/silica, 18% stearyl alcohol, 70% starch in granular form.
- Opacifier : Water based monostyrene latex mixture, sold by BASF Aktiengesellschaft under the tradename Lytron 621.
- SRP 1 : Anionically end capped poly esters.
- SRP 2 : Diethoxylated poly (1,2 propylene terephthalate) short block polymer.
- QEA : bis((C<sub>2</sub>H<sub>5</sub>O)(C<sub>2</sub>H<sub>4</sub>O)<sub>n</sub>)(CH<sub>3</sub>) -N<sup>+</sup>-C<sub>6</sub>H<sub>12</sub>-N<sup>+</sup>-(CH<sub>3</sub>) bis((C<sub>2</sub>H<sub>5</sub>O)-(C<sub>2</sub>H<sub>4</sub>O))<sub>n</sub>, wherein n = from 20 to 30.
- PEI : Polyethyleneimine with an average molecular weight of 1800 and an average ethoxylation degree of 7 ethyleneoxy residues per nitrogen.
- SCS : Sodium cumene sulphonate.
- HMWPEO : High molecular weight polyethylene oxide.
- PEGx : Polyethylene glycol, of a molecular weight of x .
- PEO : Polyethylene oxide, with an average molecular weight of 5,000.
- TEPAE : Tetraethylenepentaamine ethoxylate.

### Example 1

The following high density laundry detergent compositions were prepared according to the present invention :

	I	II	III	IV	V	VI
LAS	8.0	8.0	8.0	2.0	6.0	6.0
TAS	-	0.5	-	0.5	1.0	0.1
C46(S)AS	2.0	2.5	-	-	-	-
C25AS	-	-	-	7.0	4.5	5.5
C68AS	2.0	5.0	7.0	-	-	-
C25E5	-	-	3.4	10.0	4.6	4.6
C25E7	3.4	3.4	1.0	-	-	-
C25E3S	-	-	-	2.0	5.0	4.5
QAS	-	0.8	-	-	-	-
QAS 1	-	-	-	0.8	0.5	1.0
Zeolite A	18.1	18.0	14.1	18.1	20.0	18.1

	I	II	III	IV	V	VI
Citric	-	-	-	2.5	-	2.5
Carbonate	13.0	13.0	27.0	10.0	10.0	13.0
Na-SKS-6	-	-	-	10.0	-	10.0
Silicate	1.4	1.4	3.0	0.3	0.5	0.3
Citrate	-	1.0	-	3.0	-	-
Sulfate	26.1	26.1	26.1	6.0	-	-
Mg sulfate	0.3	-	-	0.2	-	0.2
MA/AA	0.3	0.3	0.3	4.0	1.0	1.0
CMC	0.2	0.2	0.2	0.2	0.4	0.4
PB4	9.0	9.0	5.0	-	-	-
Percarbonate	-	-	-	-	18.0	18.0
TAED	1.5	0.4	1.5	-	3.9	4.2
NACA-OBS	-	2.0	1.0	-	-	-
DETPMP	0.25	0.25	0.25	0.25	-	-
SRP 1	-	-	-	0.2	-	0.2
EDDS	-	0.25	0.4	-	0.5	0.5
CFAA	-	1.0	-	2.0	-	-
HEDP	0.3	0.3	0.3	0.3	0.4	0.4
QEA	-	-	-	0.2	-	0.5
Protease	0.009	0.009	0.01	0.04	0.05	0.03
Amylase	0.002	0.002	0.002	0.006	0.008	0.008
CBD-cellulase	0.0007	0.01	0.0007	0.008	0.0008	0.001
Cellulase	-	-	0.0007	-	0.0007	0.0001
Lipase	0.006	-	-	0.01	0.01	0.01
Photoactivated bleach (ppm)	15	15	15	-	20	20
PVNO/PVPVI	-	-	-	0.1	-	-
Brightener 1	0.09	0.09	0.09	-	0.09	0.09
Perfume	0.3	0.3	0.3	0.4	0.4	0.4
Silicone antifoam	0.5	0.5	0.5	-	0.3	0.3
Density in g/litre	850	850	850	850	850	850
Miscellaneous and minors	Up to 100%					

Example 2



The following granular laundry detergent compositions of particular utility under European machine wash conditions were prepared according to the present invention :

	I	II	III	IV	V	VI
LAS	5.5	7.5	5.0	5.0	6.0	7.0
TAS	1.25	1.9	-	0.8	0.4	0.3
C24AS/C25AS	-	2.2	5.0	5.0	5.0	2.2
C25E3S	-	0.8	1.0	1.5	3.0	1.0
C45E7	3.25	-	-	-	-	3.0
TFAA	-	-	2.0	-	-	-
C25E5	-	5.5	-	-	-	-
QAS	0.8	-	-	-	-	-
QAS 1	-	0.7	1.0	0.5	1.0	0.7
STPP	19.7	-	-	-	-	-
Zeolite A	-	19.5	25.0	19.5	20.0	17.0
NaSKS-6/citric acid (79:21)	-	10.6	-	10.6	-	-
Na-SKS-6	-	-	9.0	-	10.0	10.0
Carbonate	6.1	21.4	9.0	10.0	10.0	18.0
Bicarbonate	-	2.0	7.0	5.0	-	2.0
Silicate	6.8	-	-	0.3	0.5	-
Citrate	-	-	4.0	4.0	-	-
Sulfate	39.8	-	-	5.0	-	12.0
Mg sulfate	-	-	0.1	0.2	0.2	-
MA/AA	0.5	1.6	3.0	4.0	1.0	1.0
CMC	0.2	0.4	1.0	1.0	0.4	0.4
PB4	5.0	12.7	-	-	-	-
Percarbonate	-	-	-	-	18.0	15.0
TAED	0.5	3.1	-	-	5.0	-
NACA-OBS	1.0	3.5	-	-	-	2.5
DETPMP	0.25	0.2	0.3	0.4	-	0.2
HEDP	-	0.3	-	0.3	0.3	0.3
QEA	-	-	1.0	1.0	1.0	-
Protease	0.009	0.03	0.03	0.05	0.05	0.02
Lipase	0.003	0.003	0.006	0.006	0.006	0.004
CBD-cellulase	0.01	0.005	0.005	0.005	0.01	0.008

	I	II	III	IV	V	VI
Cellulase	0.0006	-	0.0005	-	-	0.0007
Amylase	0.002	0.002	0.006	0.006	0.01	0.003
PVNO/PVPVI	-	-	0.2	0.2	-	-
PVP	0.9	1.3	-	-	-	0.9
SRP 1	-	-	0.2	0.2	0.2	-
Photoactivated bleach (ppm)	15	27	-	-	20	20
Photoactivated bleach 1 (ppm)	15	-	-	-	-	-
Brightener 1	0.08	0.2	-	-	0.09	0.15
Brightener 2	-	0.04	-	-	-	-
Perfume	0.3	0.5	0.4	0.3	0.4	0.3
Silicone antifoam	0.5	2.4	0.3	0.5	0.3	2.0
Density in g/litre	750	750	750	750	750	750
Miscellaneous and minors	Up to 100%					

### Example 3

The following detergent formulations of particular utility under European machine wash conditions were prepared according to the present invention :

	I	II	III	IV
Blown Powder				
LAS	6.0	5.0	11.0	6.0
TAS	2.0	-	-	2.0
Zeolite A	24.0	-	-	20.0
STPP	-	27.0	24.0	-
Sulfate	4.0	6.0	13.0	-
MA/AA	1.0	4.0	6.0	2.0
Silicate	1.0	7.0	3.0	3.0
CMC	1.0	1.0	0.5	0.6
Brightener 1	0.2	0.2	0.2	0.2
Silicone antifoam	1.0	1.0	1.0	0.3
DETPMP	0.4	0.4	0.2	0.4
Spray On				

	I	II	III	IV
Brightener	0.02	-	-	0.02
C45E7	-	-	-	5.0
C45E2	2.5	2.5	2.0	-
C45E3	2.6	2.5	2.0	-
Perfume	0.5	0.3	0.5	0.2
Silicone antifoam	0.3	0.3	0.3	-
Dry additives				
QEA	-	-	-	1.0
EDDS	0.3	-	-	-
Sulfate	2.0	3.0	5.0	10.0
Carbonate	6.0	13.0	15.0	14.0
Citric	2.5	-	-	2.0
QAS 1	0.5	-	-	0.5
Na-SKS-6	10.0	-	-	-
Percarbonate	18.5	-	-	-
PB4	-	18.0	10.0	21.5
TAED	2.0	2.0	-	2.0
NACA-OBS	3.0	2.0	4.0	-
CBD-cellulase	0.0005	0.01	0.005	0.01
Cellulase	0.0004	-	-	0.0008
Protease	0.03	0.03	0.03	0.03
Lipase	0.008	0.008	0.008	0.004
Amylase	0.003	0.003	0.003	0.006
Brightener 1	0.05	-	-	0.05
Miscellaneous and minors		Up to 100%		

**Example 4**

The following granular detergent formulations were prepared according to the present invention :

	I	II	III	IV	V	VI
Blown Powder						
LAS	23.0	8.0	7.0	9.0	7.0	7.0
TAS	-	-	-	-	1.0	-

	I	II	III	IV	V	VI
C45AS	6.0	6.0	5.0	8.0	-	-
C45AES	-	1.0	1.0	1.0	-	-
C45E35	-	-	-	-	2.0	4.0
Zeolite A	10.0	18.0	14.0	12.0	10.0	10.0
MA/AA	-	0.5	-	-	-	2.0
MA/AA 1	7.0	-	-	-	-	-
AA	-	3.0	3.0	2.0	3.0	3.0
Sulfate	5.0	6.3	14.3	11.0	15.0	19.3
Silicate	10.0	1.0	1.0	1.0	1.0	1.0
Carbonate	15.0	20.0	10.0	20.7	8.0	6.0
PEG 4000	0.4	1.5	1.5	1.0	1.0	1.0
DTPA	-	0.9	0.5	-	-	0.5
Brightener 2	0.3	0.2	0.3	-	0.1	0.3
Spray On						
C45E7	-	2.0	-	-	2.0	2.0
C25E9	3.0	-	-	-	-	-
C23E9	-	-	1.5	2.0	-	2.0
Perfume	0.3	0.3	0.3	2.0	0.3	0.3
Agglomerates						
C45AS	-	5.0	5.0	2.0	-	5.0
LAS	-	2.0	2.0	-	-	2.0
Zeolite A	-	7.5	7.5	8.0	-	7.5
Carbonate	-	4.0	4.0	5.0	-	4.0
PEG 4000	-	0.5	0.5	-	-	0.5
Misc (Water etc.)	-	2.0	2.0	2.0	-	2.0
Dry additives						
QAS	-	-	-	-	1.0	-
Citric	-	-	-	-	2.0	-
PB4	-	-	-	-	12.0	1.0
PB1	4.0	1.0	3.0	2.0	-	-
Percarbonate	-	-	-	-	2.0	10.0
Carbonate	-	5.3	1.8	-	4.0	4.0
NOBS	4.0	-	6.0	-	-	0.6
Methyl cellulose	0.2	-	-	-	-	-
Na-SKS-6	8.0	-	-	-	-	-

	I	II	III	IV	V	VI
STS	-	-	2.0	-	1.0	-
Culmene sulfonic acid	-	1.0	-	-	-	2.0
Protease	0.02	0.02	0.02	0.01	0.02	0.02
Lipase	0.004	-	0.004	-	0.004	0.008
Amylase	0.003	-	0.002	-	0.003	-
CBD-cellulase	0.0005	0.01	0.0005	0.01	0.01	0.0005
Cellulase	-	-	0.0005	0.0007	-	0.0008
PVPVI	-	-	-	-	0.5	0.1
PVP	-	-	-	-	0.5	-
PVNO	-	-	0.5	0.3	-	-
QEA	-	-	-	-	1.0	-
SRP 1	0.2	0.5	0.3	-	0.2	-
Silicone antifoam	0.2	0.4	0.2	0.4	0.1	-
Mg sulfate	-	-	0.2	-	0.2	-
Miscellaneous and minors			Up to 100%			

### Example 5

The following nil bleach-containing detergent formulations of particular use in the washing of coloured clothing were prepared according to the present invention :

	I	II	III
<b>Blown Powder</b>			
Zeolite A	15.0	15.0	-
Sulfate	-	5.0	-
LAS	3.0	3.0	-
DETPMP	0.4	0.5	-
CMC	0.4	0.4	-
MA/AA	4.0	4.0	-
<b>Agglomerates</b>			
C45AS	-	-	11.0
LAS	6.0	5.0	-
TAS	3.0	2.0	-

	I	II	III
Silicate	4.0	4.0	-
Zeolite A	10.0	15.0	13.0
CMC	-	-	0.5
MA/AA	-	-	2.0
Carbonate	9.0	7.0	7.0
Spray-on			
Perfume	0.3	0.3	0.5
C45E7	4.0	4.0	4.0
C25E3	2.0	2.0	2.0
Dry additives			
MA/AA	-	-	3.0
Na-SKS-6	-	-	12.0
Citrate	10.0	-	8.0
Bicarbonate	7.0	3.0	5.0
Carbonate	8.0	5.0	7.0
PVPVI/PVNO	0.5	0.5	0.5
Protease	0.03	0.02	0.05
Lipase	0.008	0.008	0.008
Amylase	0.01	0.01	0.01
CBD-cellulase	0.0005	0.01	0.005
Cellulase	0.0008	0.001	-
Silicone antifoam	5.0	5.0	5.0
Sulfate	-	9.0	-
Density (g/litre)	700	700	700
Miscellaneous and minors		Up to 100%	

Example 6

The following detergent formulations were prepared according to the present invention :

	I	II	III	IV
Base granule				
Zeolite A	30.0	22.0	24.0	10.0
Sulfate	10.0	5.0	10.0	7.0
MA/AA	3.0	-	-	-

	I	II	III	IV
AA	-	1.6	2.0	-
MA/AA 1	-	12.0	-	6.0
LAS	14.0	10.0	9.0	20.0
C45AS	8.0	7.0	9.0	7.0
C45AES	-	1.0	1.0	-
Silicate	-	1.0	0.5	10.0
Soap	-	2.0	-	-
Brightener 1	0.2	0.2	0.2	0.2
Carbonate	6.0	9.0	10.0	10.0
PEG 4000	-	1.0	1.5	-
DTPA	-	0.4	-	-
Spray On				
C25E9	-	-	-	5.0
C45E7	1.0	1.0	-	-
C23E9	-	1.0	2.5	-
Perfume	0.2	0.3	0.3	-
Dry additives				
Carbonate	5.0	10.0	18.0	8.0
PVPVI/PVNO	0.5	-	0.3	-
Protease	0.03	0.03	0.03	0.02
Lipase	0.008	-	-	0.008
Amylase	0.002	-	-	0.002
CBD-cellulase	0.01	0.0005	0.01	0.0005
Cellulase	0.0002	0.0005	-	-
NOBS	-	4.0	-	4.5
PB1	1.0	5.0	1.5	6.0
Sulfate	4.0	5.0	-	5.0
SRP 1	-	0.4	-	-
Suds suppressor	-	0.5	0.5	-
Miscellaneous and minors		Up to 100%		

Example 7

The following granular detergent formulations were prepared according to the present invention :

	I	II	III
Blown Powder			
Zeolite A	20.0	-	15.0
STPP	-	20.0	-
Sodium sulfate	-	-	5.0
Carbonate	-	-	5.0
TAS	-	-	1.0
LAS	6.0	6.0	6.0
C68AS	2.0	2.0	-
Silicate	3.0	8.0	-
MA/AA	4.0	2.0	2.0
CMC	0.6	0.6	0.2
Brightener 1	0.2	0.2	0.1
DETPMP	0.4	0.4	0.1
STS	-	-	1.0
Spray On			
C45E7	5.0	5.0	4.0
Silicone antifoam	0.3	0.3	0.1
Perfume	0.2	0.2	0.3
Dry additives			
QEA	-	-	1.0
Carbonate	14.0	9.0	10.0
PB1	1.5	2.0	-
PB4	18.5	13.0	13.0
TAED	2.0	2.0	2.0
QAS	-	-	1.0
Photoactivated bleach	15 ppm	15 ppm	15 ppm
Na-SKS-6	-	-	3.0
Protease	0.03	0.03	0.007
Lipase	0.004	0.004	0.004
Amylase	0.006	0.006	0.003
CBD-cellulase	0.0005	0.01	0.005
Cellulase	0.0002	-	-
Sulfate	10.0	20.0	5.0
Density (g/litre)	700	700	700



Miscellaneous and minors

Up to 100%

Example 8

The following detergent formulations were prepared according to the present invention :

	I	II	III
Blown Powder			
Zeolite A	15.0	15.0	15.0
Sulfate	-	5.0	-
LAS	3.0	3.0	3.0
QAS	-	1.5	1.5
DETPMP	0.4	0.2	0.4
EDDS	-	0.4	0.2
CMC	0.4	0.4	0.4
MA/AA	4.0	2.0	2.0
Agglomerate			
LAS	5.0	5.0	5.0
TAS	2.0	2.0	1.0
Silicate	3.0	3.0	4.0
Zeolite A	8.0	8.0	8.0
Carbonate	8.0	8.0	4.0
Spray On			
Perfume	0.3	0.3	0.3
C45E7	2.0	2.0	2.0
C25E3	2.0	-	-
Dry Additives			
Citrate	5.0	-	2.0
Bicarbonate	-	3.0	-
Carbonate	8.0	15.0	10.0
TAED	6.0	2.0	5.0
PB1	14.0	7.0	10.0
PEO	-	-	0.2
Bentonite clay	-	-	10.0
Protease	0.03	0.03	0.03

	I	II	III
Lipase	0.008	0.008	0.008
CBD-cellulase	0.005	0.01	0.0005
Cellulase	-	0.0007	0.001
Amylase	0.01	0.01	0.01
Silicone antifoam	5.0	5.0	5.0
Sulfate	-	3.0	-
Density (g/litre)	850	850	850
Miscellaneous and minors		Up to 100%	

Example 9

The following detergent formulations were prepared according to the present invention :

	I	II	III	IV
LAS	18.0	14.0	24.0	20.0
QAS	0.7	1.0	-	0.7
TFAA	-	1.0	-	-
C23E56.5	-	-	1.0	-
C45E7	-	1.0	-	-
C45E3S	1.0	2.5	1.0	-
STPP	32.0	18.0	30.0	22.0
Silicate	9.0	5.0	9.0	8.0
Carbonate	11.0	7.5	10.0	5.0
Bicarbonate	-	7.5	-	-
PB1	3.0	1.0	-	-
PB4	-	1.0	-	-
NOBS	2.0	1.0	-	-
DETPMP	-	1.0	-	-
DTPA	0.5	-	0.2	0.3
SRP 1	0.3	0.2	-	0.1
MA/AA	1.0	1.5	2.0	0.5
CMC	0.8	0.4	0.4	0.2
PEI	-	-	0.4	-
Sulfate	20.0	10.0	20.0	30.0

	I	II	III	IV
Mg sulfate	0.2	-	0.4	0.9
Protease	0.03	0.03	0.02	0.02
Amylase	0.008	0.007	-	0.004
Lipase	0.004	-	0.002	-
CBD-cellulase	0.0005	0.01	0.0005	0.005
Cellulase	-	-	0.003	0.001
Photoactivated bleach	30 ppm	20 ppm	-	10 ppm
Perfume	0.3	0.3	0.1	0.2
Brightener 1/2	0.05	0.02	0.08	0.1
Miscellaneous and Minors			up to 100%	

Example 10

The following liquid detergent formulations were prepared according to the present invention (Levels are given in parts per weight) :

	I	II	III	IV	V
LAS	11.5	8.8	-	3.9	-
C25E2.5S	-	3.0	18.0	-	16.0
C45E2.25S	11.5	3.0	-	15.7	-
C23E9	-	2.7	1.8	2.0	1.0
C23E7	3.2	-	-	-	-
CFAA	-	-	5.2	-	3.1
TPKFA	1.6	-	2.0	0.5	2.0
Citric (50%)	6.5	1.2	2.5	4.4	2.5
Ca formate	0.1	0.06	0.1	-	-
Na formate	0.5	0.06	0.1	0.05	0.05
SCS	4.0	1.0	3.0	1.2	-
Borate	0.6	-	3.0	2.0	2.9
Na hydroxide	5.8	2.0	3.5	3.7	2.7
Ethanol	1.75	1.0	3.6	4.2	2.9
1,2 Propanediol	3.3	2.0	8.0	7.9	5.3
Monoethanolamine	3.0	1.5	1.3	2.5	0.8
TEPAE	1.6	-	1.3	1.2	1.2
Protease	0.03	0.01	0.03	0.02	0.02

	I	II	III	IV	V
Lipase	-	-	0.002	-	-
Amylase	-	-	-	0.002	-
CBD-cellulase	0.0005	0.01	0.005	0.01	0.005
Cellulase	0.001	-	-	0.0005	-
SRP 1	0.2	-	0.1	-	-
DTPA	-	-	0.3	-	-
PVNO	-	-	0.3	-	0.2
Brightener 1	0.2	0.07	0.1	-	-
Silicone antifoam	0.04	0.02	0.1	0.1	0.1
Miscellaneous and water		Up to 100%			

Example 11

The following liquid detergent formulations were prepared according to the present invention (Levels are given in parts per weight) :

	I	II	III	IV
LAS	10.0	13.0	9.0	-
C25AS	4.0	1.0	2.0	10.0
C25E3S	1.0	-	-	3.0
C25E7	6.0	8.0	13.0	2.5
TFAA	-	-	-	4.5
APA	-	1.4	-	-
TPKFA	2.0	-	13.0	7.0
Citric	2.0	3.0	1.0	1.5
Dodecenyl / tetradecenyl succinic acid	12.0	10.0	-	-
Rapeseed fatty acid	4.0	2.0	1.0	-
Ethanol	4.0	4.0	7.0	2.0
1,2 Propanediol	4.0	4.0	2.0	7.0
Monoethanolamine	-	-	-	5.0
Triethanolamine	-	-	8.0	-
TEPAE	0.5	-	0.5	0.2
DETPMP	1.0	1.0	0.5	1.0
Protease	0.02	0.02	0.01	0.008
Lipase	-	0.002	-	0.002

75

	I	II	III	IV
Amylase	0.004	0.004	0.01	0.008
CBD-cellulase	0.0005	0.0008	0.003	0.002
Cellulase	-	0.0007	0.0007	-
SRP 2	0.3	-	0.3	0.1
Boric acid	0.1	0.2	1.0	2.0
Ca chloride	-	0.02	-	0.01
Brightener 1	-	0.4	-	-
Suds suppressor	0.1	0.3	-	0.1
Opacifier	0.5	0.4	-	0.3
NaOH up to pH	8.0	8.0	7.6	7.7
Miscellaneous and water	Up to 100%			

Example 12

The following liquid detergent formulations were prepared according to the present invention (Levels are given in parts per weight) :

	I	II	III	IV
LAS	25.0	-	-	-
C25AS	-	13.0	18.0	15.0
C25E3S	-	2.0	2.0	4.0
C25E7	-	-	4.0	4.0
TFAA	-	6.0	8.0	8.0
APA	3.0	1.0	2.0	-
TPKFA	-	15.0	11.0	11.0
Citric	1.0	1.0	1.0	1.0
Dodecenyl / tetradecenyl succinic acid	15.0	-	-	-
Rapeseed fatty acid	1.0	-	3.5	-
Ethanol	7.0	2.0	3.0	2.0
1,2 Propanediol	6.0	8.0	10.0	13.0
Monoethanolamine	-	-	9.0	9.0
TEPAE	-	-	0.4	0.3
DETPMP	2.0	1.2	1.0	-
Protease	0.08	0.02	0.01	0.02
Lipase	-	-	0.003	0.003

	I	II	III	IV
Amylase	0.004	0.01	0.01	0.01
CBD-cellulase	0.0003	0.0006	0.004	0.003
Cellulase	0.0003	-	-	-
SRP 2	-	-	0.2	0.1
Boric acid	1.0	1.5	2.5	2.5
Bentonite clay	4.0	4.0	-	-
Brightener 1	0.1	0.2	0.3	-
Suds suppressor	0.4	-	-	-
Opacifier	0.8	0.7	-	-
NaOH up to pH	8.0	7.5	8.0	8.2
Miscellaneous and water	Up to 100%			

Example 13

The following liquid detergent compositions were prepared according to the present invention (Levels are given in parts by weight) :

	I	II
LAS	27.6	18.9
C45AS	13.8	5.9
C13E8	3.0	3.1
Oleic acid	3.4	2.5
Citric	5.4	5.4
Na hydroxide	0.4	3.6
Ca Formate	0.2	0.1
Na Formate	-	0.5
Ethanol	7.0	-
Monoethanolamine	16.5	8.0
1,2 propanediol	5.9	5.5
Xylene sulfonic acid	-	2.4
TEPAE	1.5	0.8
Protease	0.05	0.02
CBD-cellulase	0.0003	0.0006
Cellulase	-	0.0002
PEG	-	0.7

	I	II
Brightener 2	0.4	0.1
Perfume	0.5	0.3
Miscellaneous and water	Up to 100%	

Example 14

The following granular fabric detergent compositions which provide "softening through the wash" capability were prepared according to the present invention :

	I	II
C45AS	-	10.0
LAS	7.6	-
C68AS	1.3	-
C45E7	4.0	-
C25E3	-	5.0
Coco-alkyl-dimethyl hydroxy-ethyl ammonium chloride	1.4	1.0
Citrate	5.0	3.0
Na-SKS-6	-	11.0
Zeolite A	15.0	15.0
MA/AA	4.0	4.0
DETPMP	0.4	0.4
PB1	15.0	-
Percarbonate	-	15.0
TAED	5.0	5.0
Smectite clay	10.0	10.0
HMWPEO	-	0.1
Protease	0.02	0.01
Lipase	0.02	0.01
Amylase	0.03	0.005
CBD-cellulase	0.01	0.0009
Cellulase	0.005	0.001
Silicate	3.0	5.0
Carbonate	10.0	10.0
Suds suppressor	1.0	4.0

	I	II
CMC	0.2	0.1
Water/minors	Up to 100%	

Example 15

The following rinse added fabric softener compositions were prepared according to the present invention :

	I	II
DEQA (2)	20.0	20.0
CBD-cellulase	0.001	0.001
Cellulase	-	0.0005
HCL	0.03	0.03
Antifoam agent	0.01	0.01
Blue dye	25ppm	25ppm
CaCl <sub>2</sub>	0.20	0.20
Perfume	0.90	0.90
Miscellaneous and water	Up to 100%	

Example 16

The following fabric softener and dryer added fabric conditioner compositions were prepared according to the present invention :

	I	II	III	IV	V
DEQA	2.6	19.0	-	-	-
DEQA(2)	-	-	-	-	51.8
DTMAMS	-	-	-	26.0	-
SDASA	-	-	70.0	42.0	40.2
Stearic acid of IV=0	0.3	-	-	-	-
Neodol 45-13	-	-	13.0	-	-
Hydrochloride acid	0.02	0.02	-	-	-
Ethanol	-	-	1.0	-	-
CBD-cellulase	0.0001	0.001	0.0005	0.005	0.0003
Cellulase	0.001	-	0.0007	-	0.0003



	I	II	III	IV	V
Perfume	1.0	1.0	0.75	1.0	1.5
Glycoperse S-20	-	-	-	-	15.4
Glycerol monostearate	-	-	-	26.0	-
Digeranyl Succinate	-	-	0.38	-	-
Silicone antifoam	0.01	0.01	-	-	-
Electrolyte	-	0.1	-	-	-
Clay	-	-	-	3.0	-
Dye	10ppm	25ppm	0.01	-	-
Water and minors	100%	100%	-	-	-

Example 17

The following laundry bar detergent compositions were prepared according to the present invention :

	I	II	III	VI	V	III	VI	V
LAS	-	-	19.0	15.0	21.0	6.75	8.8	-
C28AS	30.0	13.5	-	-	-	15.75	11.2	22.5
Na Laurate	2.5	9.0	-	-	-	-	-	-
Zeolite A	2.0	1.25	-	-	-	1.25	1.25	1.25
Carbonate	20.0	3.0	13.0	8.0	10.0	15.0	15.0	10.0
Ca Carbonate	27.5	39.0	35.0	-	-	40.0	-	40.0
Sulfate	5.0	5.0	3.0	5.0	3.0	-	-	5.0
TSP	5.0	-	-	-	-	5.0	2.5	-
STPP	5.0	15.0	10.0	-	-	7.0	8.0	10.0
Bentonite clay	-	10.0	-	-	5.0	-	-	-
DETPMP	-	0.7	0.6	-	0.6	0.7	0.7	0.7
CMC	-	1.0	1.0	1.0	1.0	-	-	1.0
Talc	-	-	10.0	15.0	10.0	-	-	-
Silicate	-	-	4.0	5.0	3.0	-	-	-
PVNO	0.02	0.03	-	0.01	-	0.02	-	-
MA/AA	0.4	1.0	-	-	0.2	0.4	0.5	0.4
SRP 1	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3
Amylase	-	-	0.01	-	-	-	0.002	-
Protease	-	0.004	-	0.003	0.003	-	-	0.003

	I	II	III	VI	V	III	VI	V
Lipase	-	0.002	-	0.002	-	-	-	-
CBD-cellulase	.0008	.0003	.0002	.0003	.0003	.0002	.0005	.0005
Cellulase	-	-	-	.0005	-	.0002	.0004	-
PEO	-	0.2	-	0.2	0.3	-	-	0.3
Perfume	1.0	0.5	0.3	0.2	0.4	-	-	0.4
Mg sulfate	-	-	3.0	3.0	3.0	-	-	-
Brightener	0.15	0.1	0.15	-	-	-	-	0.1
Photoactivated bleach (ppm)	-	15.0	15.0	15.0	15.0	-	-	15.0

Example 18

The following pre- or post treatment compositions were prepared in accord with the present invention :

	I	II	III	IV
DEQA (2)	-	-	20.0	20.0
CBD-cellulase	0.0008	0.002	0.001	0.001
Cellulase	0.0005	-	0.0005	-
HCL	-	-	0.03	0.03
Antifoam agent	-	-	0.01	0.01
Blue dye	25ppm	25ppm	25ppm	25ppm
CaCl <sub>2</sub>	-	-	0.20	0.20
Perfume	0.90	0.90	0.90	0.90
Water / minors	Up to 100%			

CLAIMS

1. A modified enzyme comprising a catalytically active amino acid sequence of a cellulolytic enzyme linked to an amino acid sequence comprising a cellulose binding domain having a relative binding constant for binding to amorphous cellulose higher than 2.4l/gcellulose, preferably higher than 3.5l/gcellulose, more preferably higher than 4l/gcellulose.
2. A modified enzyme according to claim 1 wherein said catalytically active amino acid sequence derives from a cellulolytic enzyme having a specific activity above 50 CEVU/mg.
3. A modified enzyme according to claims 1 to 2 wherein the catalytically active amino acid sequence derives from a cellulolytic enzyme deriving from a fungal strain, preferably from a *Humicola Insolens* strain.
4. A modified enzyme according to claim 3 wherein the said catalytically active amino acid sequence derives from a ~43kD endoglucanase derived from *Humicola insolens*, DSM 1800 strain.
5. A modified enzyme according to any of the preceding claims wherein the amino acid sequence comprising a cellulose binding domain is derived from a fungal or bacterial strain, preferably a bacterial strain.
6. A modified enzyme according to claim 5 wherein said amino acid sequence comprising a cellulose binding domain is selected from the cellulose binding domains of family II and/or family IV.
7. A modified enzyme according to claim 6 wherein said amino acid sequence comprising a cellulose binding domain is selected from the family IV cellulose binding domain, preferably derived from organisms selected from the group consisting of *Cellulomonas fimi*, *Clostridium cellulolyticum*, *Myxococcus xanthus*, *Streptomyces reticuli*, *Clostridium stecorarium* (NCIMB11754) and/or *Thermomonospora fusca*.

8. A modified enzyme according to claim 7 wherein said amino acid sequence comprising a cellulose binding domain is selected from the N-terminal CBDs from *Cellulomonas fimi*  $\beta$ -1,4-glucanase CenC, CBD-dimer from *Clostridium stecorarium* (NCIMB11754) XynA and/or the CBD E3 from *Thermonospora fusca*.
9. A modified enzyme according to any of the preceding claims wherein the amino acid sequence comprising a cellulose Binding Domain is further characterised by having a relative binding constant for binding bacterial microcrystalline cellulose lower than 1l/gcellulose, preferably lower than 0.5l/gcellulose.
10. A modified enzyme according to claim 9 wherein the amino acid sequence comprising a cellulose binding domain is further characterised by having a relative binding constant for binding to amorphous cellulose to a relative binding constant for crystalline cellulose ratio above 3.
11. A modified enzyme according to any of the preceding claims providing an enzymatic hydrolysis activity on phosphoric acid-swollen cellulose higher than 60 moles glucose equivalents released per minute and per mole enzyme.
12. A modified enzyme according to any of the preceding claims further comprising a linking region between the catalytically active amino acid sequence of a cellulolytic enzyme and the amino acid sequence comprising a cellulose binding domain.
13. A modified enzyme according to claim 12 wherein the linking region is an amino acid linking region.
14. A modified enzyme according to claim 12 wherein the linking region is a non-amino acid linking region, preferably a polymer selected from PEG(NPC)2, (NH2)2-PEG, t-BOC-NH-PEG-NH2, MAL-PEG-NHS, and/or VS-PEG-NHS polymers.

15. A modified enzyme according to any of the preceding claims wherein said catalytically active amino acid sequence of a cellulolytic enzyme is alkaline.
16. A laundry detergent and/or fabric care composition comprising a laundry detergent and/or fabric care ingredient and a modified enzyme according to claims 1-15.
17. A laundry detergent and/or fabric care composition according to claim 16 wherein said modified enzyme is present at a level of from 0.0001% to 2%, preferably from 0.0001% to 0.5%, more preferably from 0.0005% to 0.1% pure modified enzyme by weight of total composition.
18. A laundry detergent and/or fabric care composition according to any of the preceding claims further comprising a builder component, preferably a zeolite A and/or sodium tripolyphosphate.
19. A laundry detergent and/or fabric care composition according to any of the preceding claims further comprising a smectite clay.
20. A laundry detergent and/or fabric care composition according to any of the preceding further comprising one or more cationic surfactants.
21. A laundry detergent and/or fabric care composition according to claim 20 wherein the cationic surfactant comprises two long alkyl chain lengths.
22. A laundry detergent and/or fabric care composition according to any of the preceding claims further comprising a dye transfer inhibiting polymer.
23. A laundry detergent and/or fabric care composition according to any of the preceding claims which is in the form of an additive, a pre-treatment, a post-treatment, a soaking treatment and/or a rinsing treatment composition.
24. A method comprising the step of contacting a fabric with a laundry detergent and/or fabric care composition according to claims 16-23 for fabric care, including anti-wrinkle, anti-bobbling and anti-shrinkage

properties to fabrics, for static control, fabric softness, colour appearance and fabric anti-wear properties and benefits.

25. A method comprising the step of contacting a fabric with a laundry detergent and/or fabric care composition according to claims 16-23 for fabric cleaning and/or fabric stain removal and/or fabric whiteness maintenance.

# INTERNATIONAL SEARCH REPORT

Inter. Appl. No.

PCT/US 99/09409

## A. CLASSIFICATION OF SUBJECT MATTER

IPC 6 C12N9/42 C11D3/386 D06M16/00

According to International Patent Classification (IPC) or to both national classification and IPC

## B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 6 C11D C12N D06M

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	<p>DATABASE WPI Section Ch, Week 9427 Derwent Publications Ltd., London, GB; Class D16, AN 94-222333 XP002090939 &amp; JP 06 158097 A (KAO CORP), 7 June 1994 (1994-06-07) abstract</p> <p style="text-align: center;">-/-</p>	1-25



Further documents are listed in the continuation of box C.



Patent family members are listed in annex.

### \* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search

5 August 1999

Date of mailing of the international search report

13/08/1999

Name and mailing address of the ISA

European Patent Office, P.B. 5818 Patentlaan 2  
NL - 2280 HV Rijswijk  
Tel. (+31-70) 340-2040, Tx. 31 651 epo nl,  
Fax: (+31-70) 340-3016

Authorized officer

Lejeune, R

# INTERNATIONAL SEARCH REPORT

International Application No

PCT/US 99/09409

## C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	<p>TOMME P ET AL: "Interaction of polysaccharides with the N-terminal cellulose-binding domain of Cellulomonas fimi CenC. 1. Binding specificity and calorimetric analysis." BIOCHEMISTRY, vol. 35, no. 44, 5 November 1996 (1996-11-05), pages 13885-13894, XP002090935 cited in the application abstract table 1</p>	1-15
A	<p>TOMME P ET AL: "Comparison of a fungal (family I) and bacterial (family II) cellulose-binding domain." JOURNAL OF BACTERIOLOGY, vol. 177, no. 15, August 1995 (1995-08), pages 4356-4363, XP002090936 abstract table I</p>	1-15
A	<p>MAGLIONE G ET AL: "properties of a genetically reconstructed prevotella ruminicola endoglucanase." APPLIED AND ENVIRONMENTAL MICROBIOLOGY, vol. 58, no. 11, November 1992 (1992-11), pages 3593-3597, XP002090937 abstract</p>	1-15
A	<p>WO 94 23113 A (GENENCOR INT ;CLARKSON KATHLEEN A (US); LARENAS EDMUND A (US); WEI) 13 October 1994 (1994-10-13) abstract</p>	16-25
A	<p>WO 97 01629 A (NIELSEN JACK BECH ;NOVONORDISK AS (DK); TIKHOMIROV DMITRY FEODOROV) 16 January 1997 (1997-01-16) cited in the application abstract page 4, line 14 - line 22</p>	16-25
A	<p>WO 95 02675 A (JEFFREYS BRIAN ;CONVENTS ANDRE CHRISTIAN (BE); TIKHOMIROV DMITRY F) 26 January 1995 (1995-01-26) abstract see examples</p>	16-25
A	<p>WO 92 17572 A (GENENCOR INT) 15 October 1992 (1992-10-15) abstract</p>	16-25
A	<p>WO 93 22414 A (GENENCOR INT) 11 November 1993 (1993-11-11) abstract</p>	16-25



# INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/US 99/09409

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
JP 6158097 A	07-06-1994	NONE	
WO 9423113 A	13-10-1994	EP 0692041 A	17-01-1996
WO 9701629 A	16-01-1997	AU 6298896 A	30-01-1997
		EP 0835302 A	15-04-1998
WO 9502675 A	26-01-1995	AU 7069294 A	13-02-1995
		BR 9407066 A	12-03-1996
		CN 1129011 A	14-08-1996
		CZ 9600110 A	12-06-1996
		EP 0708819 A	01-05-1996
		FI 960132 A	11-03-1996
		JP 9500667 T	21-01-1997
WO 9217572 A	15-10-1992	US 5246853 A	21-09-1993
		CA 2107208 A	30-09-1992
		EP 0580719 A	02-02-1994
		FI 934083 A	17-09-1993
		JP 6506359 T	21-07-1994
		US 5475101 A	12-12-1995
		US 5525507 A	11-06-1996
		US 5753484 A	19-05-1998
		US 5650322 A	22-07-1997
		CA 2107206 A	30-09-1992
		EP 0577722 A	12-01-1994
		FI 934084 A	17-09-1993
		JP 2654562 B	17-09-1997
		JP 6506829 T	04-08-1994
		WO 9217574 A	15-10-1992
		US 5328841 A	12-07-1994
WO 9322414 A	11-11-1993	AU 4232493 A	29-11-1993
		CA 2134442 A	11-11-1993
		EP 0638118 A	15-02-1995
		FI 945121 A	31-10-1994
		JP 7506394 T	13-07-1995
		US 5688290 A	18-11-1997